

Another Step Down for British Coal

THE dispute between the National Coal Board and the miners, which has brought the coal industry to a stop for the past month and which now threatens to cause havoc throughout manufacturing industry (and which may yet delay the publication of this issue of *Nature*), is yet another landmark in the decline of the British coal industry. Since the Second World War, the industry has been under severe economic pressure and it is no great surprise that there should be conflicts of interest between the miners, who rightly complain that their rates of pay have not kept pace with inflation and increased prosperity in other industries, and the National Coal Board, which remains as closely wedded as was Lord Robens to the view that the coal industry should not be allowed to decline too quickly.

The most probable outcome is that the miners will get at least a part of what they are asking for but that, now or later, many of them will lose their jobs as the coal board responds to higher costs by closing uneconomic pits. Although the government has proclaimed throughout the past few weeks that it is not a party to the dispute, there is, of course, no doubt that the conflict has been brought to a head because of the government's determination to keep wage increases in the current year within the eight per cent limit which appears to be prudent on economic grounds and, in the circumstances, it cannot entirely wash its hands of responsibility for finding a solution. At the tactical level, it should surely in the past two weeks have been more subtle in its dealings with the parties to the dispute—a determination to practise *laissez faire* economics does not preclude conversation with the coal-mining industry. It is also clear that the government has woken up rather late in the day to the way in which shortages of coal can threaten supplies of electricity. But at the strategic level, there is also a need that the government should do much more to develop a framework for energy policy in Britain.

The historic decline of the coal industry is by now a textbook example of how strictly economic pressures can affect the viability of an industry. Gone are the days when British collieries produced 210 million tons of coal or more each year. Since 1957, output has fallen steadily and, in the financial year which ended in March 1971, production amounted to a mere 142 million tons, more than half of which was used for electricity in power stations. Since 1957, the number of miners on the coal board's books has also decreased steadily, from more than 700,000 to rather less than 300,000 at the latest count. This, of course, implies a memorable increase of productivity—in 1957, each employee contributed to the production of an average of 300 tons of coal, but in 1971, the average output was 495 tons per miner per year. (The National Coal Board is more used to quoting figures for productivity in terms of the output of each face-worker during an average shift, which has nearly doubled since 1957, but such a comparison is obviously misleading when much of the improvement has been brought about

by a concentration of effort on the more productive collieries and by capital investment in machinery, which requires that larger proportions of the labour force should be engaged on maintenance.)

The number of collieries has also been sharply reduced, from more than 800 in 1957 to just about 300 in the past year, as uneconomic pits have been abandoned. With the passage of time, there have also been sharp increases of costs. To be sure, the cost of wages has been comparatively stable over the past few decades—in 1957, wages accounted for £2.41 per ton of coal, compared with £2.86 per ton of coal in 1971, but overhead costs have risen dramatically as the amounts of capital equipment to be amortized have grown. In 1957, wages accounted for 59 per cent of the total cost of mining coal, but by 1971, the wages bill was merely 46 per cent of the total, and this tendency should continue and if anything accelerate. The result is that the coal board has been compelled sharply to increase the selling price of its products which have increased from an average of £4 per ton in 1957 to £5.8 per ton in the most recent year. The miners' complaint is best illustrated by the comparatively modest pace at which average earnings in the coal industry have grown from £15.25 per week in 1957 to £28.89 per week (allowances such as concessionary coal included). The coal board's dilemma is that if there is now a substantial increase of price to meet a larger wages bill, the price of coal will increase to such a point that many traditional markets will shrink much faster than they have been doing in recent years.

In the general run of industry, consumption of coal has fallen from 73 million tons a year in 1957 to 28 million tons in the year which ended in March 1971, while in the past three years even the consumption of coal in power stations has been declining as petroleum, natural gas and nuclear power have been increasingly used for manufacturing electricity. The coal board's fear, legitimately enough, is that these tendencies will be accelerated when a settlement is reached.

What should the National Coal Board aim for in circumstances like these? A further deliberate concentration on productive pits seems now inevitable, and nobody will be surprised if output in the financial year which begins in March declines to 120 million tons or thereabouts—as it happens, the forward estimate made by the Ministry of Power (as it used to be) for 1975. The demand for coal in general industry will probably now decline more quickly than the government then estimated, while the experience of the past few years has shown that domestic demand has tailed off even more quickly than was estimated four years ago. The echoes of Lord Robens's protests against the decline of coal notwithstanding, the coal board should embrace this prospective change without too much complaint. Its chief concern should be to soften the damage which a further reduction of output will inflict on the mining communities. It should also vigorously investigate the potential benefits of further



mechanization, for even though only a tiny percentage of British coal is now mined with pick and shovel, there may be great economic benefits to be won by the replacement of machinery at present in use by equipment that is more efficient. At the same time, the coal board should safeguard the future production of metallurgical coal and the other by-product industries in which coal is used as a chemical and not merely as a source of heat, for there is at present no cause to fear that these uses are in jeopardy (and no harm would come about if there were now to be a substantial increase in the price of coal and coke for steel blast furnaces). Looking further ahead, the National Coal Board should begin to plan the kind of contraction of the industry that would be necessary if output were to fall below 100 million tons of coal a year, presumably on the basis of a still smaller labour force.

The government has other jobs to tackle. Although it may be right to resist the notion that coal should be permanently subsidized by other fuels, there is a case for asking that purchases of coal by the electricity generating industry should be planned two or three years in advance on a pattern that will help diminish the social problems in mining communities. Given that the National Coal Board and the Central Electricity Generating Board are supposedly autonomous and quasi-commercial organizations, only the government can effectively intervene. Second, the government should urgently set out to assess the impact of the discoveries of natural gas and more recently petroleum in the North Sea on the British fuel economy. The use of natural gas has already helped to provide a degree of flexibility in the generating industry which has been immensely valuable in the past few weeks of stress, but it is high time that there was an objective assessment of the potential contribution of these new resources not merely to the physical problem of generating enough electricity to provide for the country's needs but also of the potential influence on costs. Similarly, with nuclear power, the time is ripe for a serious economic study of the immediate prospects. At the very least, it would be good to know why the Central Electricity Generating Board is making such heavy weather of its plans for installing more advanced types of reactors and whether some of the potential benefits of the first fast reactors may not be won in the 1970s. One way and another, it now seems clear that there is bound to be not merely an increase of the price of coal but of energy as well—already the Electricity Council is rumbling on about higher prices, while the OPEC consortium of oil-producing countries can be relied upon (in its own best interests) to see that petroleum prices rise quite steeply in the years immediately ahead. In other words, the dispute between the miners and the National Coal Board is only one of many pressures likely to bring about a sharp increase in the price of energy in the early 1970s. It is in everybody's interests that the government should give some thought to the questions which are raised.

The condition of the British coal industry is also good grist for the mills of the conservationists, for it is a powerful illustration of how mere arithmetical calculations of the extent of a natural resource are almost irrelevant to practical questions. Britain, after all, is not physically short of coal. There is plenty beneath the ground. Moreover, as uneconomic collieries are put out of action, substantial quantities of coal are for practical purposes subtracted from the physical reserve. The seams which

these discarded collieries were intended to exploit have become uneconomic and will probably remain so. To be sure, if Britain were a developing country, or even if the economic conditions of the 1920s could be recreated, these reserves would be economically accessible. In present circumstances, however, what matters is not whether there is coal beneath the ground but whether it can be extracted at a price that potential customers are prepared to pay. The chances are that customers will be unwilling to dig deeper in their pockets for the privilege of using coal, so that the miners' entirely sympathetic demand for more take-home pay is yet another economic incentive for the substitution of other fuels for coal. But if one of the results is also a general increase in the cost of energy, the result in due course will undoubtedly be a more prudent use of energy throughout the economy, which means that annual consumption will tail off. In short, the present dilemma in the British coal industry is a telling proof that the real questions to be answered in calculating available stocks of natural resources are economic and not geophysical.

100 Years Ago



AERIAL NAVIGATION IN FRANCE*

THERE has been a most interesting sitting at the Academy of Sciences, at which M. Dupuy de Lôme read a report on his newly tried and apparently successful system for steering air balloons. M. de Lôme is one of the most eminent—if he is not the most eminent—of living French engineers. He was the first to apply steam to ships of war, and he was one of the earliest designers of ironclad frigates. The piercing of a tunnel under the English Channel is another of M. Dupuy de Lôme's long-cherished projects, and he is one of the engineers who are about to commence that gigantic enterprise. During the siege of Paris by the Prussians, M. Dupuy de Lôme offered to construct a balloon which should have steering powers of its own, and so not be totally at the mercy of the winds. That some sort of guiding power was required for the balloons which were despatched from Paris during its investment by the Germans is shown by the fact that, out of sixty balloons sent out during that period, no less than fifteen failed to carry their contents to a place of safety, some falling into the sea and several into the hands of the Prussians. After much tiresome delay, M. Dupuy de Lôme's plans were accepted by the Government of National Defence, a credit of 40,000 francs (1,600*l.*) was opened for him, and he began to construct his balloon at the Palais de l'Industrie, in the Champs Elysées. So great was the difficulty, however, in constructing an immense balloon on a totally new system, in a city completely cut off from the rest of the civilised world, that M. Dupuy de Lôme's huge machine was not ready until just four days before the capitulation. When that event took place, the balloon had to be packed up and hidden away from the prying eyes of the Germans when they partially occupied Paris. Then came the Commune, and all the disorganisation which followed. It was only after much difficulty that M. Dupuy de Lôme obtained permission to make use of the buildings of the Fort Neuf at Vincennes, whence, on the 2nd inst., he started on his trial trip.

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OLD WORLD

WATER RESOURCES

All Quiet at the Wash

WATER storage schemes in the Wash, which are now being studied in detail, could eventually yield about 600 million gallons of water a day to help meet the expected doubling in the demand for water by 2000. Decisions will be taken in 1974 on whether or not to build the first stage of the scheme which could yield 100 million gallons a day by 1983-4. These were two of the facts underlined by the Water Resources Board at a public meeting held in King's Lynn last week to discuss the proposals. The board was there in force, not merely to provide information but also to answer particular questions about the scheme.

The scheme is the result of a desk study published in 1970 which stated that with the present levels of technology it would not be possible to build a full barrage across the mouth of the Wash. It did, however, recommend that the building of four reservoirs in the Wash be studied in full. Under the proposals four bunded reservoirs would be built in stages to meet demand as and when it arose. The feasibility study will decide if the reservoirs can be built and if the benefits they bring will outweigh the disadvantages.

The possible advantages, apart from a large supply of potable water, are that valleys and agricultural land elsewhere would be saved from flooding as reservoirs, considerable recreational facilities could become available, some land could be reclaimed from the sea, and new freshwater habitats for flora and fauna would be created. The possible disadvantages are that some saltwater habitats would be lost, local fisheries will be affected, and the effects of the scheme on land drainage will have to be carefully considered.

Research into various aspects of the scheme is being undertaken by the Hydraulics Research Station, the Natural Environment Research Council and the East Anglia Economic Planning Board, all of whom were at the meeting to answer questions. The consulting engineers on the scheme, Binnie and Partners, were also represented by Mr T. G. Hammond, who explained that in the next year a trial embankment will be built at a cost of £1 million to study the best means of building the reservoir walls, and that later a second wall will be built off-shore for further studies, before the decision to go ahead is taken. Assurances were given at the meeting that all the tidal and siltation aspects of the schemes are being carefully studied, and that the effects of the scheme on the local

fisheries will also be taken into account. The meeting was surprisingly uneventful, with the audience taking the board's assurances without question; indeed many of the people packed into the Guildhall of St George seemed in favour of the scheme going ahead. Assurances were also given that the visual aspects of the scheme would be carefully studied and that the building of the scheme in stages—with the smallest of the reservoirs being built first—would allow the full effects of the scheme to be predicted before it was all irrevocably completed.

The total cost of the study will be in the order of £2.5 million and will establish the feasibility and desirability of all stages of the scheme. It is unlikely, however, unless demands for water rise faster than expected, that more than one of the four reservoirs will be built this century.

On the evidence of the public meeting it would appear that the Water Resources Board's proposals for the Wash are likely to go through much more quietly than the proposals for the scheme at Morecambe Bay, which have met with severe opposition in some quarters. The full feasibility study for Morecambe Bay, which was commissioned in 1966, has now been completed and is due for publication at the end of this month.

SCIENCE POLICY

Royal Society Puts Case

It is unusual for the president of the Royal Society to give evidence before a select committee of the House of Commons but, for the scientific community in Britain, these are unusual times. It was with this background that Sir Alan Hodgkin appeared before the Science and Technology Select Committee last week to give the Royal Society's views on the proposed reorganization.

Sir Alan expressed the Royal Society's opposition to Lord Rothschild's proposals for reforming research and development in Britain on the grounds that Rothschild's report was a "misleading oversimplification which does not provide a satisfactory framework for government research and development". Sir Alan therefore placed the Royal Society behind Sir Frederick Dainton and the Council for Scientific Policy in opposition to Lord Rothschild.

In a memorandum produced for the benefit of the Select Committee—the habit seems to be contagious—the Royal Society questions the wisdom of having had an enquiry into such an all embracing subject carried out by one scientist. The Royal Society considers that an "impression is conveyed that

the report had to be prepared in haste with little opportunity for consulting the scientific community". Sir Alan in evidence amplified this theme by saying that he did not think that "working lunches and conversations in pubs" constituted sufficient consultation when such far reaching and important changes were being considered. Sir Alan thought that a much better way of studying proposed changes would have been to set up an enquiry such as was done under Sir Burke Trend in the early 1960s where the processes of decision making were more open.

In one respect Sir Alan disagreed with Sir Frederick Dainton's report. Sir Alan told the Select Committee that he did not think that the Board of the Research Councils, if set up as recommended by the Council for Scientific Policy, should have the executive members of the research councils as members. This, however, did not imply that the Royal Society is in favour of setting up the board and the society reserves judgment on this until the future of the research councils is known.

Sir Alan admitted that Lord Rothschild had taken no apparent heed of a memorandum on the future of the research councils sent to him by the Royal Society in July 1971. This memorandum, which expressed contrasting views to those put forward by Lord Rothschild, has apparently suffered the same fate as the advice given by Sir Frederick Dainton to Lord Rothschild last summer before the green paper was published.

The Royal Society in its memorandum stresses, as did Sir Alan in evidence, that the prime factor in determining the allocation of funds for science should be the welfare of the nation, and that "responsibility and accountability should be clearly defined and allocated". Sir Alan, like Sir Frederick Dainton two weeks ago, felt that the research councils were accountable for their programmes because they produced comprehensive annual reports. Sir Alan also considered that the research councils were more accountable for their research than government departments by virtue of the reports they produced—thus throwing a new light on the problem of accountability.

While considering that the Rothschild enquiry has not been "an adequate basis for change" the Royal Society also feels that the time is not apt for a new enquiry. It does, however, recommend interim action for the government to consider, which is very similar to that suggested by the Council for Scientific Policy. The basis of the proposal is that there should be a partnership between the user ministries and the research councils in running applied research programmes. The amount of

money involved in the partnership would only be "a small fraction of that proposed by Lord Rothschild in table 4"—a sum of £7 million was mentioned by Sir Alan. The Royal Society feels that it is important to plan research on a long time scale "in order to provide stability and to ensure maintenance of a substantial core of strategic research" when applying a customer-contractor principle.

SCIENCE POLICY

Response to Rothschild

THE Science Research Council in its representations to the Secretary of State for Education and Science on the Rothschild and Dainton reports questions whether "centralization through departments or decentralization through research councils is the arrangement more likely to prove effective in managing [strategic] research intended to benefit a multiplicity of users". The SRC, although excluded from attention by Lord Rothschild, has nevertheless been examining its own programme in the light of Rothschild's proposals and in view of the government's endorsement of the customer-contractor principle.

The SRC welcomes the setting up of chief scientist's organizations in government departments and it offers to help staff them. In view of the fact that these organizations will take a few years to become effective they should be set up as soon as possible, according to the SRC. The SRC document also reiterates what its chairman, Sir Brian Flowers, has often said—that the timetable suggested by Lord Rothschild for transferring responsibility is too short for the departments to build up the necessary chief scientist organizations. The SRC has discussed the transfer of parts of its programme to the DTI and DOE and has agreed with the departments that a "proper examination of each coherent area of research . . . will take about six months". It also states that "it would be unrealistic to expect that the joint analysis of the whole programme will take much less than two or three years". The premature transfer of research programmes to the departments would, according to the SRC, undermine confidence between the research councils and government departments.

Not unsurprisingly the SRC agrees in general with Sir Frederick Dainton's proposals on the future of the research councils. The council states that the research council system as modified by Dainton "seems well adapted to ensure the effective participation of the scientific community and the users of the research . . . in the determination of programmes". The SRC has a four point plan for ensuring that the work of government departments and the re-

search councils are more closely linked.

- Each executive department should commission the applied research and development that it needs for its own purposes "or for whose potential users it is manifestly the best proxy customer".

- Members of research councils and committees should be appointed either by consultation or in part by nomination of the departments. The departments should ensure that one member of each committee should carry a departmental brief.

- The departments should be represented on the proposed Board of the Research Councils.

- Departments should use existing machinery of government to influence allocation of resources to all research and development programmes—including that of the DES.

In an analysis of its own research programme the SRC says that neither the DTI nor the DOE has identified any single research project at present sponsored by the SRC that the departments feel was applied research and would be better sponsored by them. In spite of this the departments have expressed interest in about a third of the research carried out by the engineering board "because of its contiguity to work which the departments support". The departments and the SRC are currently carrying out a review of work in control engineering, transport, mechanical and production engineering and civil engineering in order to see whether there is any duplication of work and to see whether any work funded by the SRC would be better supported by the departments or indeed whether any of the departments' research would be better under the aegis of the SRC.

INNOVATION

Do's and Don'ts

THE truth, which parts of British industry continue to ignore, that marketing is a vital factor in exploitation of new products, is underlined again by the publication of a report, published last week, on Project Sappho (Centre for the Study of Industrial Innovation, £0.75). The report is the result of three years' work by the Science Policy Research Unit of the University of Sussex, backed by the Science Research Council. The object was to find out why some innovations fail and others succeed. Much of the study consists of a comparison of success and failure with particular products. The research team claims to have identified five factors which distinguish the successes from the failures. The successful companies are said to have a better understanding of market needs,

either by formal market research, or other means. They also sell their products better, by better advertising and by liaison with customers. Development is more efficient by those who tend to use a larger development team and to spend more money on the project, and who in any case have better contacts with the scientific community in the specific area in which they are working. One important factor seems to be the placing of overall responsibility in the hands of a more senior man with more authority than his counterpart in unsuccessful companies.

The research team is at pains to point out that these factors are not offered as a recipe for success, simply as a set of guidelines which make failure that much less likely. Of the twenty-nine pairs of innovations that the group studied in detail seventeen were drawn from the chemical and twelve from the instrument industry, and the report emphasizes that the conclusions may not apply to other fields of business.

Among the common theories of what makes for success, the group's findings support the belief that there are key men whose quality is likely to affect the outcome of the project. In both industries the business innovator—the man with overall responsibility—was more successful if he were a more senior man, although he tended to have reached his rank younger in the instrument industry than in the chemical industry; the successful business innovator also tended to have more varied experience than his less successful counterpart. The group pours cold water on some common theories of successful innovation. Size seems to make little difference to the success or failure, but the size of the research team at the peak of the research and development is critical. Similarly, the level of qualification within a research department seems unimportant—a large number of qualified scientists and engineers was no guarantee of success in the cases studied. Two Sappho studies also cast doubts on the theory that innovation is more likely to succeed if it falls within a field the company knows well, and also on the view that market pressures encourage innovation. It found no connexion between competitive pressures and success or failure.

The research team hopes to publish more case studies in due course and to extend its work to include other industries. As more information becomes available, so the accuracy and importance of these, in some ways common-sense, findings will become clearer. At present the best that can be claimed for them in practical terms is, in the words of Andrew Robertson, that "they could conceivably steer some companies away from unpromising or inappropriate ventures".

NEW WORLD

Eclipse of the Stinging Fire Ant

by our Washington Correspondent

A SIGNIFICANT victory in the Department of Agriculture's fourteen year war against the imported fire ant was won last week. A scientific advisory committee recommended to William D. Ruckelshaus, Administrator of the Environmental Protection Agency, that the controversial pesticide known as 'Mirex' be continued to be used to control the ant in the southern United States. 'Mirex', the chief weapon used against the fire ant, has been criticized because of its persistence in the environment and its possible toxicity to animals and fish and in March last year Ruckelshaus moved to have its registration cancelled.

In support of its recommendation that Ruckelshaus reverse his execution order on the pesticide, the scientific advisory committee suggests that a programme to control the fire ant is necessary, and that 'Mirex' seems to be the least damaging weapon at hand. But the committee did not, however, give the pesticide a completely clean bill of health, suggesting that there are still some unknowns as far as its long-term toxicity is concerned, and recommended that its use should be restricted to areas where the ant is a special nuisance and where environmental damage would be minimal.

First introduced into the United States from South America in about 1920, the fire ant is objectionable chiefly because of its painful sting. It is also said to hamper agriculture because its mounds damage agricultural implements, and it now infests some 126 million acres in the south-eastern United States. It is a scavenger and a predator of other insects, killing both pests and beneficial species, but it is not a major pest of either plants or animals.

The campaign, which was originally aimed at eradicating the fire ant in the southern states, has, however, come up against considerable opposition both from environmental groups and from scientists. The chief charge levelled against the eradication programme was that the fire ant is not a major pest species and, although a nuisance, it does not merit the concentrated attack it has received and the consequent environmental damage that has been caused. In 1967, a committee of the National Research Council concluded that eradication of the fire ant is not biologically feasible, and suggested that even if eradication were feasible it would not be desirable. The issue finally came

to a head when the Environmental Defense Fund sought a court injunction last year to prevent the Department of Agriculture from using 'Mirex' in an attempt to eradicate the insect. The injunction was denied, but the Department of Agriculture was forced to abandon its goal of eradication and instead concentrated on a control programme, avoiding application of 'Mirex' to estuarine and forested regions.

The EPA's decision to cancel the registration of 'Mirex' was taken on the basis of doubts concerning the safety of the pesticide and its persistence in the environment. The Allied Chemical Corporation, exercising its rights under the Federal Insecticide, Fungicide and Rodenticide Act, however, asked for a scientific review of the pesticide, and it is the fruit of that which has produced the recommendation to Ruckelshaus that the use of 'Mirex' be continued.

The committee, of which the chairman was Dr C. H. Van Middellem of the University of Florida, bases its recommendation on the fact that no instances of acute toxicity have been reported, there is no evidence of damage to vegetation and there is no significant build up of the pesticide in the human food chain. Moreover, the committee states "most investigations of the effect of 'Mirex' on invertebrates and vertebrates in the natural habitat have failed to demonstrate any significant changes in their populations", but there has been evidence of 'Mirex' residue build-up in crustaceans and in some vertebrates that are predatory on ants. As for chronic toxicity, the committee suggests that insufficient studies have been made, and although 'Mirex' has been found to be carcinogenic in mice when fed in high doses, "no conclusions can be reached concerning the carcinogenicity of 'Mirex' for man until it has been studied in other mammalian species".

The committee therefore concludes that use of 'Mirex' is justified since it is not a proven health hazard and because environmental damage seems to be relatively insignificant. The clear inference is that until the hazards are fully evaluated by future chronic toxicity studies, the public can justifiably be exposed to the pesticide.

The Department of Agriculture's plans for the use of 'Mirex' to control the fire ant were made clear in the Administration's budget message. They entail a level of effort in 1973 that is essentially the same as the present year,

with some 20 million acres being treated with 'Mirex' bait, at a cost of about \$7 million. Mr Leo Iverson, of the Animal and Plant Health Service, who is in charge of the Federal programme, said last week that if use of 'Mirex' is denied, the programme would have to rely on less effective pesticides and that these may be more damaging to the environment.

ENVIRONMENT

Pollution Tax

by our Washington Correspondent

PRESIDENT NIXON last week sent to Congress his third and so far his most modest message on the environment. A declaration of the Administration's intent to introduce a variety of legislation to curb pollution and protect the environment, the message included two measures that have grabbed the headlines—a tax on emission of sulphur dioxide from smokestacks and a ban on poisoning predators on Federal lands—but few other far-reaching measures.

In his message to Congress, President Nixon called his proposed legislation a plan to build on the base of environmental legislation that he has already introduced and which is at present mostly bottled up in Congressional committees or awaiting final passage through the Congressional mill. The controversial water pollution control legislation, which was introduced by the Administration last year, and which has since been considerably toughened by Senator Muskie's committee in the Senate and by the House Public Works committee, is a case in point.

The item in the proposed legislation that is likely to spark off the most discussion and the most opposition from industry is the long-awaited proposal to impose a tax on the emission of sulphur dioxide by factories. Billed as a measure that will ensure "application of the principle that the costs of pollution should be included in the price of the product", the proposed tax is, however, much less ambitious than the measure touted by the Administration in last year's environmental message and again when President Nixon transferred to Congress the second annual report of the Council on Environmental Quality. Environmentalists are already accusing the Administration of bending to industrial pressure by weakening the legislation in election year, but the very fact that President Nixon has proposed taxing sulphur emissions at all is a bold stroke. Even his most ardent critics

acknowledge that the proposition embodied in legislative proposals will give welcome public ventilation to the arguments that surround an emissions tax.

In short, an emissions tax is a device to make it more costly for a company to carry on polluting than to introduce controls. The idea is to charge a polluter a fixed tax for every pound of sulphur he emits as sulphur dioxide, thereby increasing the prices of his products if he chooses to pass the price on to the consumer and leaving him at a competitive disadvantage, or decreasing his profits if he chooses to absorb some of the tax. And the most discussed concept is the introduction of a sliding tax which could be imposed almost immediately and increased yearly until a maximum is reached in about 1976, thus steadily increasing the incentive to curb sulphur emission.

But the Administration's draft bill is neither so simple or so tough. What is being proposed is that a sulphur tax of 15c a pound should be imposed in 1976, with no sliding scale leading up to that figure. Moreover, the Administration plans only to tax polluters in areas which do not meet the standards prescribed by the Clean Air Act. Polluters in areas which meet the secondary standards of the Clean Air Act would be charged at the rate of 10c a pound, while those in areas which meet both the primary and secondary standards would have to pay nothing for their sulphur pollution.

Such a graded emissions tax is consistent with the Administration's thesis that pollution costs should be reflected in product prices. But some environmentalists have been quick to suggest that the proposed bill would be more a method of enforcing the Clean Air Act than of taxing polluters. Congressman Les Aspin, who has already introduced his own legislation to impose a sliding emissions tax, for example, has suggested in a letter to President Nixon that the Administration's proposal could lead to some subtle and undesirable results. He suggested, for example, that large corporations with plants both in heavily polluted and in unpolluted areas would simply use low grade fuel in the clean area and high grade fuel in the more polluted area, thereby continuing to pollute the environment with sulphur dioxide. He also suggested that a sliding tax, introduced in the near future, at a relatively low level would be more of an incentive than a tax to be imposed in 1976 at the maximum level.

But for their part, members of the Administration argue that it is justifiable to hit polluters in heavily polluted areas harder than those in clean areas since the marginal cost of their pollution is greater. Another suggestion is

that differential taxation may encourage polluting plants to move away from heavily polluted areas. But environmentalists counter that simply transferring pollution is not the same thing as reducing it.

Environmentalists have not, however, shown a lifelong devotion to the pollution tax. When the measure was first suggested, the common reaction was that a tax would simply be a licence to pollute, and that companies would merely pass the costs on to the consumer and carry on polluting. But over the past year organized environmental groups have gradually come to see a pollution tax as a strong incentive to institute abatement measures. They also cite the fact that most industrialists have declared their opposition to the measure as proof that it would force them to take the painful steps to reduce their sulphur emissions. A measure of the environmentalists' interest in the concept is the fact that in August last year a number of groups joined to form a "Coalition to Tax Pollution", and set up an office on Capitol Hill.

If a sulphur tax is imposed, one inevitable result will be price increases, with electricity prices being among the most hard pressed. Although such a consequence would simply be a reflexion of the fact that social costs would then be paid for in hard cash, supporters of the legislation will clearly have to do a good public relations job to convince the consumer of the benefit of paying higher bills. The coming Congressional debate on the Administration's proposal will at least help to clarify some of the issues and consequences of fiscal means to control the use of environmental resources.

Other, less controversial measures outlined in President Nixon's environmental blueprint for 1972 include:

- Controls on the disposal of toxic wastes on land and in underground waters.
- A ban on the poisoning of predators on Federal lands. This measure will chiefly benefit the coyote which some observers have suggested is in danger of being driven to extinction by past methods of wholesale killing. The ban was suggested to the Council on Environmental Quality by a task force from the University of Michigan Institute for Environmental Quality which, under the chairmanship of Stanley A. Cain, has recently presented a weighty report on predator control to the CEQ.
- The initiation of a programme aimed at integrated pest management to be conducted by the Department of Agriculture, the National Science Foundation and the Environmental Protection Agency.
- Expansion of programmes to test children in high risk areas for lead concentrations from lead based paint.

TECHNOLOGY ASSESSMENT

OTA Lives On

by our Washington Correspondent

NOBODY can seriously claim that the House of Representatives is a technologically competent body in its own right. With only one scientifically qualified member in its ranks, it has to rely greatly on outsiders and on the work of its committee staff for advice on legislation that has a bearing on science and technology. That is the chief reason why the House of Representatives last week took a step designed to strengthen its scientific advisory system by agreeing to a bill to set up an Office of Technology Assessment. The bill was passed by a margin of 256 to 117, and now goes to the Senate where the Rules Committee has already scheduled hearings for March 2.

A suggestion which was first mooted by Emilio Q. Daddario when he was chairman of the House Subcommittee on Research and Development, the Office of Technology Assessment is designed to give independent analyses of technological matters being considered by Congress. It will be especially concerned with looking out for important consequences of technological legislation in areas such as the environment and employment, and its supporters claim that it would have been especially valuable in helping Congress to come to the correct decision on the SST. Underlying the debate on the necessity for the office is the idea that Congress is unable at present to match the technological expertise of the Administration, and needs its own think tank to be able effectively to challenge the Administration's technical decisions.

If the bill is passed essentially unchanged by the Senate, the Office of Technology Assessment would work chiefly by contracting out studies to universities or non-profit organizations. Studies could be initiated by committee chairmen or at the request of the ranking minority member or a majority of members of any Congressional committee. It would be managed by a director and a board of management, which would also have the authority to initiate technological assessment studies and which would be responsible only to Congress. It will be given \$5 million to get established in its first two years of operation, and may cost about \$7 or \$8 million a year after that. Its full staff complement is expected to be between 50 and 100 people.

But not every member of the House is convinced that Congress needs an Office of Technology Assessment to pinpoint the likely consequences of its technological actions. Mr H. R. Gross of Iowa, for example, termed the proposed office a "boondoggle" during the

debate in the House last week, and suggested that it would not save much taxpayers' money.

What seemed to bother most of the opponents of the Bill last week was that the functions that are envisaged for the office could be carried out either by committee staff or by expanding the staff of the General Accounting Office or the Library of Congress. Few congressmen, however, questioned the need for better and more broadly based studies of technological matters in Congress, and there was not much discussion of such important questions as whether the tools of technology assessment are yet sufficiently refined to justify setting up such an office. Moreover, experience in other countries of attempts to use such assessment studies as a basis for legislation has not been completely faultless. Perhaps the most ambitious technological assessment study, and one which nicely exposes the problems of such an exercise, was the effort of the Roskill Commission which spent two years and £2.1 million in choosing the best site for London's third airport, only to have its recommendation promptly rejected by the British Government.

Nevertheless, the legislation to set up the Office of Technology Assessment survived the House debate essentially intact. The only change that was forced was in the constitution of the management board, which instead of consisting of eleven members (four appointed by the President, two by the House, two by the Senate, the comptroller general, the director of congressional research and the director of the Office of Technology Assessment) would now consist of ten members who would come in equal numbers from the House and the Senate. The amendment which forced the change was proposed by Mr Jack Brooks of Texas to ensure that the office will be responsible only to Congress—Mr Brooks pointed out that under the original proposal, a majority of board members would be presidential appointees. Perhaps it is a measure of the interest of members of Congress in technological affairs that only 48 were present to vote on the amendment.

SACCHARIN

Off the List

by our Washington Correspondent

WITH the bitter taste left by the cyclamates farce still in its mouth, the Food and Drug Administration must have received with some trepidation the news that a study in Wisconsin has come up with tentative evidence of bladder tumours in rats fed on a diet heavy with saccharin. Although the findings are preliminary and the tumours are not necessarily cancerous, the FDA's

response was to take saccharin off the list of food additives generally recognized as safe (GRAS), and to take action on an eighteen month old recommendation from a committee of the National Academy of Sciences by suggesting that the average daily intake of the sweetener should be limited to one gram per person (about 60 tablets, or seven 12 oz. bottles of so-called diet drink).

The FDA's action touched off barely a ripple of public anxiety, and thankfully was not followed by the precipitate worldwide action that greeted the tentative finding in 1969 that cyclamates produced bladder cancers when fed in huge quantities to rats over a period of two years. But if the Wisconsin experiment does produce evidence that the bladder tumours in rats fed with saccharin over a long period of time are cancerous, then the Delaney Amendment to the food and drug laws requires that saccharin, too, must be taken off the market. The Delaney Amendment disallows the use of any substance as a food additive if it is found to cause cancer when fed to animals.

The tentative findings on saccharin were made in the laboratories of the Wisconsin Alumni Research Foundation (WARF), through a study supported by the Sugar Research Foundation. Although no details of the experiments have been released, it is understood that the tumours were found in three out of twenty rats fed a diet of 5 per cent saccharin during a two-year feeding experiment. The experiment has not yet run its full length, and the suspected tumours have still to be examined by pathologists to see if they exhibit signs of malignancy. The WARF experiment is only one of about twelve two-year feeding experiments now being conducted, and the FDA says that none of the other experiments has so far produced any "adverse findings".

Nobody can accuse the FDA of precipitate action in its handling of saccharin; in fact, quite the reverse. In July 1970, the *ad hoc* subcommittee on non-nutritive sweeteners of the National Academy of Sciences recommended that saccharin be taken off the GRAS list, and that a safe usage level would be about 5 mg/kg/day (equivalent to about 0.25 to 0.35 grams per day for an adult). The committee based that recommendation on the results of several experiments which indicated that no effect on the growth, survival, production of bladder stones, kidney pathology or bone marrow hyperplasia are produced when rats are fed saccharin at the rate of one per cent of the daily diet.

The committee did point out, however, that at higher daily dose levels, notably at five per cent of the diet, saccharin does have some adverse effect on each of these variables, and sug-

gested that one per cent of the daily diet could be taken as the level at which saccharin had no effect. On the basis that safe levels in man are conventionally set at one hundredth of the no-effect level in animals, the committee came up with the recommendation that saccharin intake should be limited to 5 mg/kg/day, but suggested that this limit is probably unduly conservative. But the FDA considered that the suggested level was too low and asked the committee to reconsider its recommendations in the light of its general conclusion that "present and projected usage of saccharin in the United States does not pose a hazard". This the committee duly did, and came up with the suggestion that 15 mg/kg/day would be a "safe" level (equivalent to about 1 g for an adult).

The Food and Drug Administration then allowed almost a year to elapse before taking action on the committee's suggestion that saccharin should be taken off the GRAS list. On June 25, 1971, the FDA published in the Federal Register the proposal to remove saccharin from the list and to set the new limits on its use recommended by the NAS committee. Such proposals are usually open to comment for thirty days before being put into effect, but in this case the FDA allowed another seven months to go by and was finally rushed into taking action when it received the preliminary WARF findings.

Although the delay would be difficult to justify on most criteria, at least it allowed the dust to settle over the cyclamate ban and perhaps forestalled a panic reaction to tentative results. But, on the other hand, if the FDA had proceeded with reasonable speed on the NAS recommendations, it would have taken action against the sweetener not on the basis of its possible carcinogenicity, but because of its other proven chronic effects when fed to rats. The public could then have been spared the suggestion that saccharin may cause bladder cancers until sufficient evidence is available to form a balanced conclusion. All the feeding studies should be completed by the end of 1972.

The possibility that saccharin may cause bladder cancer in rats fed massive doses of the sweetener also adds a final note of irony to the farce that surrounded the removal from the market of cyclamates in 1969. The key experiment that led to the ban consisted of feeding rats not pure cyclamate, but a mixture of cyclamate and saccharin in a 10:1 proportion (*Science*, **167**, 1131; 1970). The FDA then assumed that it was the cyclamate that caused the cancer, not the saccharin or a reaction between the two. The finding now that saccharin is suspected of causing identical bladder tumours is ironic to say the least.

SCIENCE POLICY

Pressgang or Partnership?

This article on the Rothschild and Dainton reports by Sir Gordon Cox, formerly secretary of the Agricultural Research Council, is based on a public lecture given in the University of Leeds on February 8, 1972.

It is now more than half a century since a report of a committee on the machinery of government (cmd. 9230) drew attention to the great and increasing importance of scientific research and intelligence in public affairs and recommended two parallel and complementary developments. First, for conducting enquiries and research for general use of administrative departments but under the supervision of authorities not charged with administrative duties, and second, for conducting enquiries and research of more particular kinds under the supervision of administrative departments.

In the case of the first, the committee envisaged an organization with "a primary duty . . . to keep in touch with scientific workers in various fields throughout the country . . . for the benefit of the administrative departments as a whole", a concept which led to the establishment of the research councils. As to the second the committee said that a minister in charge of an administrative department must have under his control an organization to keep him in touch with relevant scientific knowledge and to undertake "competent, swift and self-contained" enquiries to enable him to deal with specific executive problems.

How successful have these two developments been? On the first the Trend Committee was quite unequivocal—"It is a striking tribute to the prescience of the Haldane Committee that the basic principle of the autonomous research council which it originally sponsored should have stood the test of time so successfully and that, when the need for adjustment became apparent, the nature of the essential changes required should appear to be on lines which the committee itself foresaw". And in the same year, speaking at the 50th anniversary dinner of the MRC, Lord Hailsham (Mr Quintin Hogg as he then was), Minister for Science, paid a glowing tribute to the work of the research councils and the administrative foundations on which their work had been built; he emphatically rejected the idea, which had recently been put forward, that the MRC should be handed over to the Department of Health and Social Security (DHSS) "which would have the double effect of

divorcing it from other scientific work, and subordinating it to the requirements of day-to-day administration".

It is rather more difficult to assess the development of scientific services in the executive departments, largely because many of their activities are not open and accountable to the scientific world in the same way as those of the research councils; and the observed end results depend on factors other than the scientific ones. Thus a proposal by the Milk Marketing Board in 1955 to do some small-scale experiments on cross-breeding from French Charolais cattle was approved by the MAFF six years later; again, at the beginning of the century progressive farmers believed, from their experiments, that bulls were substantially more efficient as beef producers than steers, but until 1971 the ministry could not relax the bull licensing regulations which prevented the testing and exploitation of the findings of those pioneers. In these and similar cases the outsider can only observe the overall results, and cannot assert that the scientific processes involved inside the ministry were not "competent and swift". Now, however, Lord Rothschild tells us that, in the headquarters of some ministries at least, the scientific organization does fall short of what he believes to be necessary.

Nevertheless the message seems to be that the research councils are doing things that they ought not to do; that they are not doing things that they ought to do; that even if they are doing the "right" things a large proportion could be done better by executive ministries; that their priorities are wrong; and finally that resources are being diverted to the councils that could be used better for other, non-scientific, purposes.

This last point is different in kind from the rest, because while it is fair enough to criticize the councils if they mismanage the resources allotted to them, it is quite unfair to saddle them with responsibility for political judgments made by others. In his Royal Society of Arts lecture (December 8, 1971) Lord Rothschild is reported to have said that the scientific establishment should ponder whether the money for the proposed high flux beam reactor "whatever that is" could not be better spent on, for example, four hospitals, or four prisons,

or Titian's Death of Actaeon, or 200,000 television sets for the old and house-bound. This remark suggests that the scientists are selfishly hogging resources (in this case for a purpose which Lord Rothschild has apparently not taken the trouble to understand) that they ought to know are needed more urgently elsewhere; it comes nearer to the language of the demagogue than to that expected from the head of the government's Central Policy Review Unit. Lord Rothschild is a dab hand at trailing red herrings, but whatever changes he may propose in machinery of government he must not be allowed to obscure the fundamental fact that the distribution of available resources between the Science Vote and the other votes is a matter for political decision. All sorts of pressures influence the decision, but once the Science Vote has been allocated it is the duty of those who account for it to spend it only on activities within their remit, though a hundred Actaeons should die.

Of course, there has been plenty of criticism of research councils before. *Pace* Professor Waddington, with his delightful picture of the secretaries as renaissance Popes, neither the chief executive nor the members of their councils regard themselves as infallible; they know only too well how far short of perfection their endeavours fall, and they would accept some of the criticism as just. But they would also say, I think, that much of it stems from muddled thinking or worse.

Possibly this view is held more widely, and accounts for the absence from the Rothschild report of any specific criticism and for the fact that the reasons for "the proposal to transfer the ARC to MAFF" referred to by Dainton have never been exposed to public view. Or, maybe, there were no reasons, but only old-fashioned emotions; it would have to be admitted that the agricultural research service has impregnated the MAFF with a good many seminal ideas, but in these permissive times that is scarcely a good enough reason for a shot-gun marriage.

Emotion does indeed peep out in Rothschild's references to the "haves" in the research councils and the "have nots" in the departments, a strange comparison when we remember that the research councils' budgets are equivalent to a per cent or so of that of the departments, and one which makes 1 *Kings*, 21, an obvious source for a prefatory quotation to enable Rothschild to keep up with the Daintons—"Ahab said to Naboth 'Give me your vineyard that I may have

it for a vegetable garden, because it is near my house'".

If there are few rational grounds for finding fault with the present system, the presumption is that the drastic changes proposed by Lord Rothschild are confidently expected to bring about an even better state of affairs. This would indeed represent the triumph of hope over experience, for in many respects the implementation of the recommendations would mean a return to arrangements that have already been tried and found wanting.

Before 1956 much agricultural research was financed by the ministry on the advice of the ARC. As Professor H. D. Kay records in his biography of Sir William Slater, "In spite of considerable opposition, but with the powerful assistance of his chairman together with the sympathy of the House of Commons Select Committee on Estimates, Slater was able, in 1954, after five years of patient and tenacious endeavour, to end this cumbersome and inefficient division of responsibility". (The chairman referred to was Lord Rothschild, who in those days seemed to hold the naive idea that farmers were the principal customers of agricultural research (see, for example, Jubilee Lecture, Long Ashton 1953).) Since that time the ARC has had to live with the consequences of its own decisions and neither it nor the country has been any the worse for that.

The nature of agricultural and medical research, carried out as it is in many institutions of different characters, makes an autocratic attitude to management impossible; equally the diversity of subject matter excludes any possibility of an authoritarian approach to policy formation, which must be based on a consensus. Even in the rare cases where an authoritarian line has to be taken, for example to veto expenditure on a half-baked proposal, the authority is not *ex officio* but derives from a consensus about standards.

In the formation of the consensus, since after all it is scientific research we are talking about, directors of research and other scientists naturally play a leading part, but (in ARC affairs, for example) farmers and officers from the ministry's various technical arms make essential contributions. Nearly half of the 200 or so people who make up the governing bodies of research institutes are farmers, while at the last count I found about 40 MAFF professional officers of one sort or another on ARC committees. It is hard to sustain the argument that the customer has no say.

The word committee is evidently somewhat of a red rag to Lord Rothschild and this is perhaps the place to nail his innuendo about the alleged inefficiency of research councils' administration. As the accounts published annually show, the headquarters costs

are very low—in the case of the ARC, for example, less than 3 per cent of the budget, even though various non-administrative items are for convenience included with central costs. A straightforward comparison with the executive ministries is not easy to make, but careful comparative study of the published estimates and accounts strongly suggests that the percentage administrative costs could run into double figures. In the 1961–62 session the Select Committee on Estimates recommended that the MAFF should undertake periodic calculations of administrative costs or, failing that, the Treasury should establish some means of future control: it would be interesting to see some of the figures that have emerged from these exercises, together with estimates of the cost of the proposed chief scientist organization.

It would not be surprising if politicians took the view that the concentration of scientific endeavour is maintained at too great a cost in relation to the other pressing needs of the community; consequently, in the jargon of the economic planners, a manpower shake-out is called for. The recipe proposed by Rothschild for this shake-out, with the customer-contractor principle as the basis for a sort of scientific selective employment tax, would create far more problems that it would solve. If indeed it is thought, as a matter of political judgment, that too high a proportion of the country's resources are going to the research councils, it would be better to say so explicitly; the scientific community would squeal, but would understand and no doubt eventually accept the argument. The present proposals are dishonest in expecting the research councils to exercise scientific leadership while depriving them of effective power and freedom of decision; the executive ministries are to choose the water and drive the scientific horse to it, leaving the research councils to persuade it to drink.

The late Sir Frederick Bawden and others have shown how slipshod the thinking behind the customer-contractor proposal is. Nevertheless, some are still saying politely that perhaps there are areas where it might work. I have yet to identify them in any biological field. Let me take one trivial, almost non-biological, example.

We all know that British horticulture is vulnerable to overseas competition and must therefore become even more efficient to survive. Less dirt on the glass means more photosynthesis inside the glasshouse and a greater yield of tomatoes. So let us place a contract for improved methods of cleaning glass. As Bawden said in relation to hard wheat, there are some questions to be asked—how clean is clean, and would the National Union of Agricultural Workers mind if the scientists recommended a

stronger solution of hydrofluoric acid than that already used by the growers—but one could no doubt frame a reasonably clear contract.

But the whole concept is wrong from the start, because the object is to get more tomatoes, and cleaner glass is only one of a number of intricately related factors bearing on the ultimate object. So what? Rothschild might say; the chief scientist's organization will analyse the problem and place a group of inter-related contracts. Would it though? Not in time to save the tomato industry, and not unless some miracle transmutes the departmental organization which so effectively discourages peripheral vision and lateral thinking. When I say I wonder where the dirt on the glasshouses comes from, I am just indulging in ivory tower evasion of the real issue—but perhaps a high proportion of the dirt does come from the growers' own chimneys, and maybe cost-benefit analysis would show that what they should go for is not more research but planning permission for higher chimneys.

The fact is that the scientific method is not yet understood in parts of Whitehall. For example, if Rothschild's "better than 90 per cent chance" of success is to be attained also in the application of experimental findings it goes without saying that one of the many necessary conditions is that the technical conclusions on which application is to be based have been validated statistically. Yet not so long ago one of the most senior persons in the MAFF, talking to farmers about changes that should be made in the experimental farms, could say "we must free our experimental development work from the encumbrance of statistical accuracy, concentrate on developing and demonstrating new systems and provide factual information on what the systems involve". If these are the standards of those who would control applied research and development, a scientist, remembering what medicine and agriculture owe to Fisher, Yates, Bradford Hill and others, can but echo the words of Naboth—"The Lord forbid that I should give you the inheritance of my fathers".

At the present time there is little doubt that to set up departmental machinery to operate the proposed contractual arrangements would be more costly, less flexible and less effective than an alternative of the Dainton type. The alternative only needs cooperation, on a basis of equality and mutual respect, between "customer" ministries and research agencies—this is perfectly possible today, with only minor adjustments of the system. Will the men from the ministry come out in the open and talk about cooperation, instead of stalking the streets by night with cloak and dagger?

Control of Strategic Research in Agriculture

This comment on the Rothschild and Dainton reports is by Dr R. D. Keynes, director of the Institute of Animal Physiology, Babraham.

In sections (ii) and (iii) of the appendix to his report, Lord Rothschild justifies his proposal that "only" 77.5 per cent of the ARC's budget should be transferred to MAFF and controlled on the customer-contractor principle on the grounds that the remaining expenditure is devoted to basic or strategic research which lacks an applied objective likely to be realized in a specified time. He arrives at the residual 22.5 per cent by adding together the budgets of the ARC's units in universities and of a few institutes that include my own. Since my autonomy would thus not be eroded, it might be imagined that I would be less disturbed than the majority of my colleagues about the probable effect of some of Lord Rothschild's propositions. This is not the case, and I wish to explain why I share their disquiet.

I should make it clear from the outset that I have no quarrel with Lord Rothschild's basic objectives if they are first to ensure that the customer has an adequate voice in the commissioning and conduct of short-term applied research, and second to strive towards a rational basis for deciding how much to spend on long-term strategic and fundamental research. Nor do I think that the existing system for determining research priorities is perfect. But as he makes very clear in his report, the identifiable imperfections are to be found more in the scientific organization—or rather lack of it—of government departments than in any obvious failure on the part of the research councils to respond to the practical needs of the nation. I therefore question the logic of handing over to MAFF responsibility for such a large part of the ARC's funds, and hence of effective control of priorities.

It is quite unrealistic to make a sharp distinction between ARC institutes and units primarily engaged on long-term strategic and fundamental research, and those, by implication, concerned mainly with short-term applied research. At all the ARC's establishments there is, as Lord Rothschild says there should be, a mix of short, medium and long-term research. As far as the proportions of the mix are concerned, the Institute of Animal Physiology does perhaps occupy

one pole of a continuum, since we are mainly occupied with long rather than short-term research aimed at achieving a better understanding of the physiology of farm animals. But it is a continuum, and even at those institutes whose objectives are most immediately realizable, some strategic research is also done. There are often joint projects that link the poles, and I understand that at the Letcombe Laboratory, one of those placed by Lord Rothschild in the same category as Babraham, there is extensive collaboration, very much at the applied level, with members of the staff of the Agricultural Development and Advisory Service of MAFF. A recent analysis of the overall deployment of the ARC's resources has shown that the percentages of its funds devoted respectively to short-term applied research, long-term strategic research on specifically agricultural processes, and fundamental research, are roughly 20, 60, 20. These proportions are the result of past decisions on research priorities, and I would certainly not regard them as sacrosanct, nor would I claim to possess a magic formula for determining precisely what they should be. But I am confident that they are more nearly correct than those indicated by Lord Rothschild's perfunctory classification of the ARC's activities. Moreover, the arguments about the indivisibility of short and long-term investigations, put forward by Professor Riley for plant breeding, apply with equal force to animal husbandry, and indeed to the whole spectrum of the research undertaken by the ARC.

The question fundamentally at issue is therefore how strategic research on food and agriculture is to be controlled so that the country gets the best value for its money. From the discussions of the past few weeks it is evident that many industrial research directors consider that the customer-contractor principle is difficult to apply in its purest form even for development programmes with well defined objectives. Those planning agricultural research face a highly complex situation. Not only must the customer represent three quite distinct and sometimes conflicting interests—the agricultural industry, the consumer of agricultural produce, and

the community at large—but cost-benefit analysis is often not directly applicable, and social benefits that are impossible to cost enter into the equation. Is a MAFF official, whose chief aim is to increase the economic efficiency of an agricultural process, the right person to decide on the degree of environmental pollution or ecological disturbance that can be tolerated as an accompaniment? And how does one decide exactly how much to spend on research on animal welfare, such as investigation of the behaviour of farm animals subjected to intensive husbandry conditions? Again a strictly cash-conscious customer is not the right arbiter. If, then, the customer-contractor principle is hard to put into effect for short-term applied research and development in agriculture, it will be doubly so for long-term strategic research.

Lord Rothschild has recently said (see *The Times*, January 19) that "There is nothing to prevent a government department commissioning relevant basic or strategic research within an applied research and development programme". As has been pointed out in *Nature*, this may indeed be claimed at the present juncture by departments interested more in empire building than in exercising their proposed new responsibilities efficiently on the lines laid down in the green paper. But the whole tenor of Lord Rothschild's arguments is surely against the appropriateness of customer control for strategic research.

A central and certainly essential feature of Lord Rothschild's recommendations is that in order to be capable of acting intelligently as customers, departments must first acquire a chief scientist and supporting staff; and as he emphasizes, MAFF only has such an organization in an embryonic form. It will take an appreciable time for it to be brought fully into operation, and to gain the confidence of the farmers on the one hand and the research workers on the other. Research budgets have to be planned well ahead. I find it hard to see how Lord Rothschild's own conditions for imposing the new regime can possibly be met as quickly as he proposes. The task of incorporating MAFF's contracts into the ARC's research programme, even on a rational and acceptable scale, will be a complicated one. I suggest that the new financial arrangements should be introduced on a gradual basis over several years, by mutual agreement between the ARC, MAFF and all the other interested parties.

A Question of Efficiency in Research

This comment on the Rothschild and Dainton reports is by Dr M. Gibbons and Dr R. D. Johnston of the Department of Liberal Studies in Science, University of Manchester.

THE stated objective of the Rothschild report is to provide a more efficient framework for the operation of government research and development. The means proposed for achieving this are summarily expressed in the government-endorsed customer-contractor principle. This principle is based on a clear distinction between basic and applied research; basic research is by nature unplannable, and should be allowed to proceed according to the dynamics of science with a minimum of external control, whereas applied research, by definition, is directed towards a specific goal which should be established under a customer-contractor relationship. Dainton has questioned the possibility, and the wisdom, of such a sharp distinction, and the important area of strategic research, and the possibility of its subversion, has been the subject of much comment.

In spite of these difficulties, however, a more fundamental question to be examined in this context is whether the specific findings of research, be it basic or applied, are the only, or even the most important, contribution of the research worker. Recent studies of the role of science and technology in the process of technological innovation would suggest that this is not the case.

Science and technology can no longer be regarded as being linearly related in the sense that "science discovers, technology applies". Rather there is much evidence to support the view that scientific and technological activities are carried out by two distinct but related communities pursuing different ends and using different means, but that each community relies for its development on a complex matrix of interactions or mutual coupling. More specifically, coupling, in the form of interaction between the generators and users of information, is "person embodied" and is optimized by close, continuing, and pluralistic collaboration between the scientific and the technological communities.

The importance of effective coupling and its contribution to innovation have

been emphasized by a number of studies in the USA, summarized by Price and Bass (*Science*, **164**, 802; 1969) and by comparable studies of innovation in British industry by Dr J. Langrish and colleagues from the University of Manchester and Dr C. Freeman and Dr A. Robertson, of the University of Sussex. This has been highlighted by Freeman who has said that "access to recent fundamental research was of critical importance in nearly half of the process innovations studied. This sometimes took the form of working more closely with outstanding consultants, sometimes of recruiting outstanding scientists from academic life or immigrant scientists, sometimes of academic scientists themselves exploiting innovations, and sometimes simply of careful study of world scientific and technical journals".

The role of various types of coupling has been further demonstrated by an on-going study of the types of information used and the ways in which it was obtained by technologists in the resolution of technical problems that arose during the development in British industry of a wide range of new products characterized by their commonplace, unspectacular nature. In particular it has been shown that the major contribution from science was in the transfer, *via* a person-to-person contact, of information which was not being specifically sought, and which was frequently not from the research front.

Though most of these findings have been based on studies of the interaction of science and technology in the industrial sphere, there is no evidence to suggest that they are not applicable also to government departments. Indeed, much of the relevant research work in the USA has been in the context of research commissioned by government agencies or departments.

The results of these studies relevant to the current debate can be summarized as follows. First, the chief contribution of science to technology occurs in the transfer of a wide variety of information on the occasion of personal contact between the two communities. Second,

the information transferred is rarely being specifically sought. Being by nature unplannable, attempts to optimize such transfers must concentrate on developing an environment favourable to coupling.

We do not suggest that the chief activity of the researcher should not be directed towards basic or applied research as needs require, or that the information obtained is not important as a contribution to the body of knowledge or in solution of a problem. The research cited above, however, does suggest that this is not the most important way in which the researcher contributes to the process.

It would seem, in the light of this evidence, that the implementation of the Rothschild recommendations may have at least two potentially adverse effects on the scientific community. First, the rigid application of the customer-contractor principle could serve to isolate scientists from technological problems as they attempt to define their areas of autonomy by retiring into basic research. Second, to use the scientists simply as a source of specific information for the solution of specific problems unnecessarily restricts the role they might play in contributing to the resolution of these and other problems, because it is clear from the studies cited above that the scientists' role in technical problem solving is by no means easily defined, even in general terms, from the outset and that more often than not they contribute in an unexpected way. In both these ways, the present proposals would have the effect, in our view, of reducing coupling and hence the efficiency which is their aim. At the same time, they include little which might promote an increase in this vital coupling.

Rothschild, in paragraph 21, does stress the importance of dialogue, but with the limited purpose of selection of applied research targets. The need, however, is for a mechanism which allows for and encourages continuing dialogue between potential users such as the chief scientist, controller research and development, and their staff, and a wide range of the scientific community engaged in both basic and applied research. Changes in the organization of government research and development should have as a chief goal the expansion and augmentation of coupling so that the benefits of science can flow more readily to the entire community.

NEWS AND VIEWS

From One Kind of Smog to Another ?

LOS ANGELES, renowned for all kinds of reasons, has in the past few years had the doubtful distinction of being the place for photochemical smog. The mechanism is simple, at least in outline. Unburned hydrocarbon molecules from motor vehicle exhausts react with ozone molecules to produce oxidizing radicals and unstable molecules which, by ill-luck, are able to turn sulphur dioxide into sulphuric acid. The result is not merely that ordinary air becomes foul and even unbreathable but that the sulphuric acid in combination with drops of water and particles of dust and smoke creates the peculiarly smoky kind of atmospheric turbidity through which the inhabitants of Los Angeles attempt to see.

There is no doubt that motor vehicles are the principal outlets for photochemical smog in large cities, for they are the sources for the unburned hydrocarbon, sulphur dioxide and also smoke. Meteorological conditions are important, for the sequence of reactions leading to photochemical smog requires the production of significant ozone concentrations sufficiently near the ground for the unburned hydrocarbons not to have been dispersed by diffusion or by wind, but even so, the formation of photochemical smog in the large conurbations of North America has been a striking proof of the formation of ozone at surprisingly low levels in the atmosphere.

The article by Atkins, Cox and Eggleton on page 372 of this issue, an analytical *tour de force* in its own right, will be a still more vivid proof of the ubiquity of ozone formation in significant concentrations and of the processes by which photochemical smog is formed. The starting point for their work is the measurement of ozone concentrations near the ground, based on the chemiluminescent reaction between ozone and ethylene. By the comparison of ozone concentrations on different days, it now seems that the background concentration when the air near the ground is easily dispersed is somewhere between 20 and 40 parts per thousand million. The same measurements have, however, shown that when meteorological conditions are suitable and in particular when the winds are light, concentrations of ozone near the ground may be three times or more as much. The demonstration that the excess ozone comes not by diffusion downwards through the troposphere but is formed locally instead rests on the correlation between concentrations and meteorological conditions.

It goes without saying that the concentrations now reported from rural Harwell are much smaller than those which have recently been carried out in Los Angeles where the greatest concentrations of ozone measured at Harwell are exceeded on half the days in the year and where concentrations of ozone as great as 580 parts per thousand million have been recorded. Even so, it is a surprise to say the least of it that the countryside in England can be thus affected. It is no surprise, when concentrations of ozone as great as 80 parts per thousand million are recorded, that unburned hydrocarbons and nitrogen oxide should conspire to oxidize sulphur dioxide to sulphuric acid.

The precise implications of these measurements for the control of photochemical smog in the conurbations where it is a public nuisance are not easy to define but there can be no doubt that the controlling element in this sequence of events is the presence of unburned hydrocarbons in motor vehicle exhausts. Certainly it will not be realistic to hope that ozone will not be formed on sunny days even in temperate latitudes. The remedy, if any, is for the designers of internal combustion engines to discover.

To Psi or not to Psi

LAST year molecular biologists on the circuit of annual meetings in the late summer and early autumn heard in no uncertain terms that Haseltine had carried out a set of experiments that, on the face of things, exploded the elegant model of the regulation of the expression of ribosomal RNA genes in *Escherichia coli*. This model had been proposed by Travers and his colleagues in 1970 (*Nature*, **228**, 748; 1970; and *Cold Spring Harbor Symp. Quant. Biol.*, **35**, 415; 1970) and has been widely espoused since. And as anybody who has read Haseltine's report in last week's issue of *Nature* (**235**, 329; 1972) will realize, this storm in a teacup stems from contradictory observations of the pattern of transcription, in cell free systems, of purified *E. coli* DNA by purified RNA polymerase in the presence of various factors.

Travers and his colleagues originally reported that virtually no ribosomal RNA is made when *E. coli* DNA is transcribed *in vitro* by RNA polymerase including sigma factor. When, however, a factor called psi (obtained either from extracts of *E. coli* or from the replicase of phage Q β , of which psi is a subunit) is added, synthesis of ribosomal RNA *in vitro* is specifically stimulated by close on two orders of magnitude. Moreover, Travers *et al.* also found that the nucleotide ppGpp prevents this specific stimulation of transcription by psi factor. This last observation seemed particularly significant because ppGpp, the so-called magic spot I, accumulates when wild type *E. coli* cells are starved of amino-acids and during such starvation the synthesis of ribosomal RNA is preferentially turned off. Travers and his colleagues not unreasonably concluded that synthesis of ribosomal RNA in *E. coli* may well be subject to positive control. Psi factor, they argued, is a positive control element which somehow allows *E. coli* RNA polymerase to initiate transcription specifically at the otherwise inaccessible promoter sites of the ribosomal RNA genes. They also envisaged that the nucleotide ppGpp somehow antagonizes psi factor, so the extent of ribosomal RNA synthesis depends at least in part on the relative concentrations of psi factor and ppGpp.

By complete contrast Haseltine reported in *Nature* last week that although psi factor stimulates the transcription of *E. coli* DNA *in vitro* this stimulation is entirely non-

specific. Using different hybridization assay procedures for the analysis of the products of *in vitro* transcription—procedures which he argues are more sensitive than those used by Travers *et al.*—he finds that in the absence as well as in the presence of psi factor between 7 and 14 per cent of the RNA made *in vitro* is ribosomal RNA. Furthermore he finds that ppGpp does not alter the proportion of ribosomal RNA made *in vitro* irrespective of the presence or absence of psi factor. While working at Harvard in the very laboratory in which Travers and his colleagues had done their experiments, Haseltine found in the deep freeze samples of RNA polymerase and a sample of DNA which Travers had used eighteen months earlier. Realizing that he was about to upset the apple cart, Haseltine used these samples, and found that in his reaction conditions the enzyme which Travers used transcribes ribosomal RNA in the absence of psi factor; he also found that one of the DNA preparations which Travers had used as template supports an anomalously low level of ribosomal RNA synthesis. This Haseltine attributes to the presence of extensive single strand regions in the DNA.

As Haseltine comments, Hussy, Pero, Shorestein and Losick have independently found that about 10 per cent of the RNA transcribed *in vitro* from *Bacillus subtilis* DNA by purified polymerase without psi factor is ribosomal RNA. Furthermore, although Pettijohn *et al.* (*J. Mol. Biol.*, **52**, 281; 1970) reported that, like Travers *et al.*, they could not detect ribosomal RNA in the products of *in vitro* transcription by RNA polymerase in the absence of psi factor, in last Wednesday's issue of *Nature New Biology* (**235**, 204; 1972) Pettijohn reports that using a more sensitive hybridization assay procedure he can detect ribosomal RNA made by RNA polymerase without added psi factor. Furthermore, he finds that 16S ribosomal RNA is made *in vitro* in the absence of psi before 23S ribosomal RNA is made and of course these two RNAs appear in this order *in vivo*.

In short, these various experiments with cell free systems seem to have led to an impasse; one set of experiments indicates psi factor is required to transcribe ribosomal RNA; the other, and larger, set indicates psi is irrelevant. In part, of course, these striking differences probably stem from differences in experimental procedures. For example, Haseltine is critical of the hybridization assay used by Travers and his associates to detect ribosomal RNA sequences in the mixture of RNAs made *in vitro*. A sensitive and reliable assay is obviously crucial and Haseltine finds the Travers assay to be subject to uncontrollable variations. Likewise, Pettijohn suggests that differences in assay procedures may be at the bottom of the current discrepancies. On the other hand it is curious, to say the least, that Haseltine, knowing his observations would be the centre of contention, should have gone to the trouble of taking out from the freezer preparations of enzyme and template used by Travers without at the same time using reaction conditions absolutely identical to those used by Travers *et al.* Haseltine says that his reaction conditions "are similar to those that have been used before", but one might well ask why they were not identical: as is well known from experiments with cell free systems which support protein synthesis, by varying the precise reaction conditions protein synthesis can be made independent of or absolutely dependent upon added initiation factors.

It is, however, sterile to pursue such arguments *ad*

nauseam, for what one is, or should be, really interested in is how ribosomal RNA synthesis is regulated in the intact cell. If, as may well be the case, this regulatory mechanism is extremely sensitive to factors such as the ionic strength and the concentration of sulphhydryl groups in the reaction mixture it simply may not reproducibly survive the abuses to which it is subjected by molecular biologists attempting to reassemble it *in vitro*. Indeed, having reached this impasse with their experiments with *in vitro* systems, those intent on elucidating the mechanism of controlled transcription of ribosomal RNA genes might well be advised to find a new approach to the problem, adopting "back to the cell" as their rallying cry. Ultimately, no matter how the controversy over the role of psi factor in transcription *in vitro* is resolved, it must be proved that psi either is or is not required *in vivo*.

Apart from getting to the bottom of the part played by the nucleotide ppGpp in the stringent-relaxed control of RNA synthesis *in vivo* the obvious approach is to search for conditional lethal mutants affecting psi factor. If Travers's ideas are correct, cells with a temperature sensitive psi factor should cease to synthesize ribosomal RNA at the non-permissive temperature, but if Haseltine is correct, ribosomal RNA synthesis should continue unabated. Whether or not such mutants can be isolated depends, among other things, on just how many essential roles psi factor plays in the cell and on that score there may be some surprises in store.—From our Cell Biology Correspondent.

Imploding-Exploding Waves

AMONG the more striking astronomical observations which have been made in recent years are the much-discussed results of Weber (see *Nature*, **235**, 199; 1972) which seem to indicate the presence of a large flux of energy in the form of gravitational waves emanating from the galactic centre. If confirmed, these observations could not fail to have considerable implications for astronomy, for they indicate the presence of a very peculiar situation at the centre of the galaxy, where space-time cannot be treated adequately in a Newtonian or special-relativistic way. Unless (and until) some newer theory of gravitation turns out to be required, it will be necessary to take the theoretical implications of general relativity much more seriously than has been done hitherto. Indeed, more analysis seems necessary within Einstein's theory in order to understand what the theoretical implications of that theory really are.

Much work has been done on the theory of black holes, which now may perhaps be considered as one of the aspects of general-relativistic physics which is better understood. It is still quite unclear, however, how black holes might be used to explain an energy flux on the scale that Weber seems to observe. Perhaps help can be sought in a much less thorough understood aspect of the theory, namely the propagation of the gravitational waves themselves, in which the non-linearities of the theory lead to self-scattering effects and even the production of space-time singularities (that is, regions in which the normal ideas of local physics would cease to apply). It is in this context that the work of Marder (see page 379 of this issue) has a particular interest.

Very few examples are known of explicit solutions of Einstein's equations which exhibit this self-scattering; and

those which are explicitly known all possess unsatisfactory features such as infinite total energy. Marder presents a class of solutions which represent waves of finite energy (initially) which implode and then re-explode. Although they still possess features which preclude them as being totally realistic models, they nevertheless seem to be worthy of study as models in which imploding gravitational waves either do, or do not, result in space-time singularities. It is to be expected that there should exist some initial criterion which, if fulfilled, would imply that the strength of the waves is sufficient that space-time singularities should be the result. Marder's solutions could well lead to some insights as to what such a criterion might be.

These imploding-exploding waves are of a toroidal character. Their only unsatisfactory feature seems to be that they do not fall off appropriately towards infinity in two directions at one advanced and one retarded time. But they could well form the basis of further work in this field.—From a Correspondent.

MUSCLE

New Twists to the Twitch

from our Molecular Biology Correspondent
REGARDLESS of the distant thunder that always seems to greet any thoroughgoing reductionism, one might hazard the view that the innermost secret of muscular contraction will be revealed only when the interaction of three species of molecules, myosin, actin and ATP, is fully understood. In the past year or two many apparently disconnected and sometimes contradictory features of this system have fallen into place, and a pattern is now beginning to take shape which invites for the first time some inferences to relate biochemical events to structural changes. The kinetic analysis of Taylor and his colleagues of the myosin ATPase and the work of Eisenberg and Moos on its activation by actin have led to a scheme that has now been tested in an important article by Lymn and Taylor (*Biochemistry*, **10**, 4617; 1971).

The earlier work showed that the rate-determining step in the myosin ATPase reaction was not the hydrolysis of the substrate but the release of the products (which defined in operational terms could of course in itself be a complex process with more than one kinetic component). This explained at once the enigmatic "early burst", consisting of a very rapid initial round of ATP hydrolysis. Lymn and Taylor have now examined the kinetics of the actomyosin complex—or rather that of actin with heavy meromyosin (or proteolytically truncated myosin), which is

more tractable in respect of its solubility characteristics. Using a stopped-flow method, they first show that the early burst persists in the actin complex with much the same rate as in uncomplexed myosin, and involves the cleavage of close enough to one molecule of ATP per site. The steady state hydrolysis rate is of course a good order of magnitude higher in actomyosin, and the effect of ATP is to dissociate the complex.

The question now is whether the dissociation precedes or follows the hydrolysis. The rate for the hydrolysis step being now known, it was necessary to assess the rate of dissociation, and this was done by stopped-flow measurements of the turbidity decrease that accompanies it. This reaction turns out to be extremely fast, and it follows that the dissociation of the actomyosin precedes the hydrolysis of ATP. To explain the ATPase activation by actin, moreover, one is now compelled to postulate that the actin promotes the dissociation of the hydrolysis products from the myosin active sites. Using their rapid column technique to separate the labelled products, phosphate and ADP from the protein, Lymn and Taylor find that this is indeed so: the elution profiles show that in the presence of actin the lifetime of the enzyme-product complex is diminished by a factor of perhaps ten, depending on the ionic strength.

One arrives therefore at a cycle of events beginning with the binding of ATP to the ATPase sites of actomyosin. This brings about the dissociation of the actomyosin; there follows the hydrolysis of the ATP and the reassociation of the actomyosin, with the contingent accelerated displacement of the hydrolytic products. Now this is the simplest possible scheme, which leaves out of account possible intermediate steps, such as conformational changes that might be expected to accompany and regulate the binding processes, and the attractive but barely supported notion of interaction between the sites on the two chains of the myosin molecule. So far as they go, however, the conclusions are firm, and in quantitative terms the measured maximum rate constants are compatible with the steady-state rate of Eisenberg and Moos.

In terms now of the structural cycle of contraction the enzymatic events fit in as follows: the arrival of ATP and its attachment to the myosin site causes the dissociation of myosin head in question from its actin subunit; this leaves the myosin head region, or bridge, free to swing, as it is known from X-ray and electron microscopy to do, into its relaxed position; the ATP is hydrolysed at this juncture, so leaving the myosin free to bind to the actin globule now nearest to it. This causes the hydrolysis products to be liberated

and the myosin head returns to its "tension" position in what is regarded as the drive stroke in the cycle, at the end of which therefore the picture has been translated one unit along the actin filament.

The first indications of the intermediate steps that might be anticipated to occur within the framework of such a basic scheme follow hot on the heels of Lymn and Taylor's study. Trentham, Bardsley, Eccleston and Weeds (*Biochem. J.*, **126**, 635; 1972) have used a kind of cascade of coupled enzymes under conditions of rapid reaction to generate at some removes a spectroscopic response to the liberation of ADP and phosphate from ATP and heavy meromyosin. The formidable difficulties of avoiding the intrusion of insufficiently rapid rate processes at points in the sequence having been overcome, the method makes possible a comparison of the dissociation rates of ADP and of phosphate ion from their preformed complexes with heavy meromyosin, with the rate of their appearance in the primary cycle of ATP hydrolysis. The dissociation rates turn out to be somewhat the faster. Thus it seems that the rate-determining step is sandwiched between hydrolysis and product release.

Another path to the same conclusion involved the use of a modified substrate, thioATP, the purine chromophore of which undergoes a spectroscopic change on binding to myosin. On mixing with heavy meromyosin a biphasic rate curve is obtained, either phase being fitted by a single rate constant. The first is presumed to represent hydrolysis. The rates of thioADP and also of ADP release from the active site were observed by chasing off the former with ATP, and the latter with thioATP, and the measured values confirm that it is not the dissociation *per se* that is the slow step. The inference again is that a slow step intervenes after hydrolysis, and most probably involves a conformational adjustment of the protein. This can be accommodated without difficulty into the cycle described by Lymn and Taylor, the complex of myosin in this putative conformational state with ADP and phosphate being the preponderant form to be found in the functioning muscle. This expanded scheme may serve also to explain a number of spectroscopic manifestations of ADP binding.

In regard to this last matter, Uchida, Tanaka and Hiratsuka (*Biochim. Biophys. Acta*, **256**, 132; 1972) have now introduced a chromophore into heavy meromyosin by modification of certain thiols. The ATPase activity is retained—indeed enhanced, as with other limited thiol modifications—and the addition of ATP or ADP generates measurable spectral shifts in the new chromophore. The profiles of the magnitudes of these

shifts as a function of nucleotide concentration are difficult to interpret with any certainty, but tend to start again the hare of a functional heterogeneity of sites—that is to say interaction between the two myosin heads.

ENVIRONMENT

Contamination by Lead

from a Correspondent

A CONFERENCE on lead in the environment which was organized by the Institute of Petroleum in association with the Chemical Society and the British Occupational Hygiene Society in London on January 27 provided a unique opportunity in Britain for a discussion on a wide range of problems related to the nature, distribution and consequences of environmental lead. The meeting was opened by Sir Eric Ashby, chairman of the Royal Commission on Environmental Pollution.

Mr R. L. Stubbs (Lead and Zinc Development Association) reviewed the importance of the various forms of lead used and pointed out that large quantities of the scrap metal are recycled. With respect to the contamination of the environment he referred in particular to lead used in gasoline, the combustion of coal and other natural products, and the release of lead during its fabrication.

Professor P. J. Lawther, Dr B. T. Commins, Dr J. McK. Ellison and Mr B. Biles (Medical Research Council's Air Pollution Research Unit, London) presented their recent results of measurements of airborne concentrations of lead, the visual appearance under the electron microscope of lead containing particles emitted from engines and information related to a limited study of the blood lead content of London taxi drivers. They found that the concentration of particulate lead in one busy street was about twice that found nine years earlier, but that measurements at a site away from a street indicated for the past six years a possible decline in airborne lead there. In another study measurements indicated that only about one third of the airborne lead at a site away from a street was attributable to the motor vehicle. An examination of the extremely small particles from engines indicated that published information on the deposition, retention and uptake of airborne lead could not be applied to these special particles.

Drs L. H. P. Jones and C. R. Clements (Grassland Research Institute) showed that the uptake of grasses grown in soils containing lead is small and concluded that man's dietary intake of lead from this source is low because animals absorb little lead from grass

and that which is absorbed accumulates in bone.

Dr H. Egan (Laboratory of the Government Chemist) presented information relating to the lead content of soil, vegetation and foodstuffs. He concluded that since 1940 there was no evidence that the amount of lead in the British diet had changed substantially.

Dr R. I. McCallum (Nuffield Department of Health, Newcastle) reviewed the modes of lead absorption and intoxication and the laboratory tests for assessing the effects on health following excessive exposure to lead. He concluded that for the general population there seemed to be no sound medical evidence to support clinical or sub-clinical lead poisoning.

Dr D. Barltrop (St Mary's Hospital, London) reviewed the environmental exposure to children and the behavioural factors which arise from excessive lead intake. He concluded for children that lead inhaled from the atmosphere is small by comparison with other sources of lead—for example, the chewing of lead containing paint and the ingestion of lead containing dust.

In the two and a half hours of general discussion the topics debated included (a) the interpretation of concentrations of lead found in the atmosphere in terms of vehicular movement and meteorological factors; (b) the financial and technological consequences of removing lead from petrol; (c) the relevance of industrially accepted exposure levels in relation to the air breathed by the general public; and (d) the need for further monitoring of the environmental lead including additional tests on the blood of British children. It was clear from the meeting that a great deal of information about lead is already known.

VIRAL TRANSFORMATION

Genetics of Control

from our Cell Biology Correspondent

THE temperature sensitive mutant strains of polyoma virus and Rous sarcoma virus which have been characterized respectively by Eckhart and Dulbecco and Martin clearly indicate that the maintenance of the transformed-cell phenotype of mammalian and avian fibroblasts transformed by these viruses depends on the continued expression of at least one viral gene. Viral transformation is not a hit and run affair, neither is the persistence of the viral genome without its partial expression sufficient to maintain transformation. It would, however, be unwise to assume from these observations that the properties which distinguish virus-transformed from untransformed cells are under the immediate control of the viral genome. Indeed Renger and Basilico (*Proc. US Nat. Acad. Sci.*, **69**, 109; 1972) have now reported an intriguing set of experiments which strongly suggest that the maintenance of several diagnostic properties of transformed cells depends on the continued expression of some cellular gene(s).

Instead of mutagenizing stocks of transforming virus before screening for temperature sensitive mutations which affect the transformed cell phenotype, Renger and Basilico took mouse 3T3 cells transformed by presumably wild type SV40 virus and, by a selective killing procedure, selected for cells which behave at 32° C as typical transformants, at least as far as cell multiplication in dense cultures is concerned, but which at 39° C behave in these conditions as untransformed cells. In other words, they selected cells from populations of transformants which

Xenon Isotopes in Natural Gas

IN next Monday's *Nature Physical Science* (February 21) M. S. Boulos and O. K. Manuel of the Nuclear Chemistry Department, University of Missouri, show from analysis of the isotopic composition of xenon found in natural gas that plutonium-244 was at one time to be found on Earth and that the isotope was probably present when the Earth was formed.

Previous analysis of natural gas samples has been unable to show unambiguously that plutonium-244 contributed to the anomalous enrichment of some of the xenon isotopes found in natural gas because a longer lived spontaneously fissioning isotope—uranium-238—also contributed to the xenon content. The work reported in this article is, however, sufficiently accurate for there to be no ambiguity,

and although uranium-238 is shown to contribute to the xenon content, the observed distribution of the xenon isotopes cannot be described solely by the fission of uranium-238 and a contribution from another spontaneously fissioning isotope—plutonium-244—is needed to explain the data.

Plutonium-244 has a half life of 6.55×10^{10} year for spontaneous fission but its decay is dominated by its 8.2×10^7 year half life for decay by alpha emission. On fissioning it produces xenon isotopes of atomic mass between 131 and 136, and these isotopes are expected to be enriched in xenon that has been in contact with plutonium over that found in atmospheric xenon. Boulos and Manuel also find that xenon-129 is enriched in their gas samples.

multiply in the face of contact inhibition at 32° C but not at 39° C.

Having isolated several such temperature sensitive cells they assayed other parameters at 32° C and 39° C. As expected, the cells, which have SV40 T antigen at both temperatures, grow to high saturation densities at 32° C but only to low saturation densities typical of untransformed cells at 39° C. At 32° C the temperature sensitive cells are able to form colonies when plated on top of confluent monolayers of untransformed 3T3 cells, but at 39° C colony formation in these conditions is severely impaired and little greater than that observed with untransformed cells. Moreover, although the temperature sensitive transformants at 32° C and 39° C require for growth much less serum than do untransformed cells, they stop making DNA in response to cell with cell contact at the higher temperature but continue to make DNA when in equally dense cultures at 32° C. Preliminary experiments also apparently indicate that the ease with which these cells can be agglutinated by wheat germ agglutinin is temperature sensitive; at 39° C they are less susceptible to agglutination than at 32° C.

One way to account for all these properties is to suggest that the transforming gene of the resident SV40 genome(s) carries a temperature sensitive mutation. This is unlikely to be the case, however, for the SV40 virus rescued when the temperature sensitive transformants are fused with permissive BSC-1 monkey cells is apparently wild type, not mutant. The temperature sensitive mutation in these cells which affects the transformed cell phenotype seems therefore to be in a cellular gene(s) and the finding that these cells cannot be retransformed at 39° C by infection with multiplicities of up to 200 SV40 particles per cell supports this view.

In the same issue of the *Proceedings* (*ibid.*, 152), Smith, Gelb and Martin report a most unexpected finding with BALB/C cells abortively transformed with SV40 virus. From DNA hybridization experiments they conclude that cells of two of three lines of abortive transformants, cells which after infection by SV40 had only transiently behaved as if transformed and had then resumed the untransformed cell phenotype, contain about five copies of the complete SV40 genome per diploid cell genome; for comparison stable SV40 BALB/C 3T3 transformants contain only two copies of SV40 DNA per cell. To convince any sceptics of the validity of their conclusion Smith *et al.* could always fuse cells of these three lines with monkey cells and show that SV40 virus can only be rescued from the two lines which contain SV40 DNA.

LYMPHOMAS

Monkey Models

from our Medical Virology Correspondent

A RECENT report from the University of California National Centre for Primate Biology (Stowell *et al.*, *Lab. Invest.*, **25**, 476; 1971) records the largest ever outbreak of simian malignant solid lymphomas in twenty-four adult rhesus monkeys. This outbreak occurred over a period of 25 months and the attack rate was eighteen cases per 1,000 adult rhesus monkeys. This astonishing incidence—the reported occurrence of malignant lymphomas in non-human primates is very rare—together with the remarkable similarity to Burkitt lymphoma in man, may provide a useful model for the experimental study of human lymphoma. There are also several other intriguing features. No single potential or actual carcinogenic factor was identified in this outbreak although three herpes virus-like agents have been isolated. Ten of the twenty-four affected monkeys, however, had been involved in experiments with malarial parasites and subjected to exposure to X-irradiation.

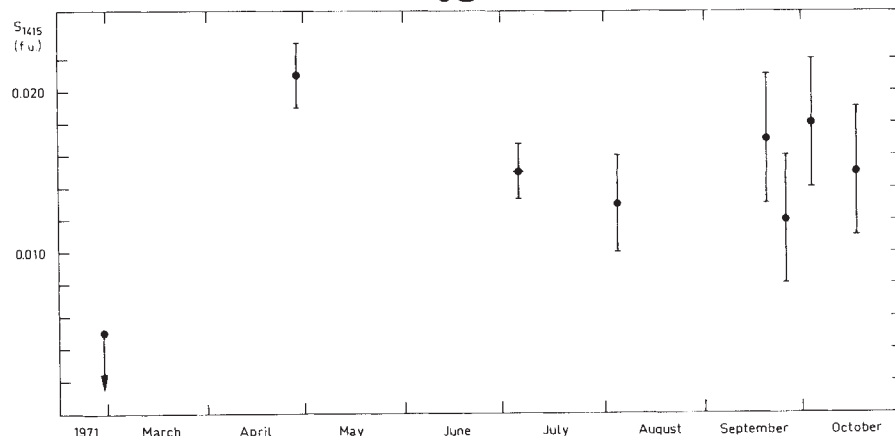
The possible relation between hepatitis associated with Australia antigen and the pathogenesis of primary liver cell cancer in man has been previously discussed (see *Nature New Biology*, **232**, 100; 1971). More recent information is now available. Persistent carriage of Australia antigen is common in patients with a defective immune response, for example, lymphocytic

leukaemia, chronic renal failure and lepromatous leprosy. There is also epidemiological and experimental evidence for immunological changes, including depression of the immune response in malaria.

A. J. Zuckerman (*Brit. Med. J.*, **1**, 49; 1972) has proposed that the high frequency of carriage of Australia antigen in the serum of apparently healthy people living in the tropics may be related to this immunological phenomenon coupled with repeated biting by blood-sucking insects. This hypothesis may be extended further because an altered immune reactivity in malaria may also affect the outcome of chronic infection with a virus by allowing the virus to become frankly oncogenic and possible by interfering with the immune reaction to neoplastic cells. Indeed, Salaman *et al.* (*J. Gen. Microbiol.*, **59**, 383; 1969) and Wedderburn (*Lancet*, **ii**, 1114; 1970) demonstrated an increased incidence of malignant lymphoma caused by murine oncogenic viruses in mice infected with *Plasmodium berghei yoelli*.

A similar relation might exist between chronic infection with the serum hepatitis virus and the subsequent development of hepatoma (A. J. Zuckerman, *loc. cit.*), and it is of interest that an association of Burkitt's tumour and holoendemic malaria has also been suggested by Kafuko *et al.* (*East Afr. Med. J.*, **46**, 414; 1969). The possibility therefore that sub-human primates might prove a useful model for the study of lymphomas, hepatoma and other malignant tumours presents an intriguing and exciting challenge.

Radio Observations of Cygnus X-1



CONTINUING observations of Cyg X-1 at radio frequencies are being carried out with the Westerbork array. The figure here shows results obtained by L. L. E. Braes and G. K. Miley during 1971, which are reported in next Monday's issue of *Nature Physical Science* (February 21).

The sudden appearance of the source in April 1971, and its subsequent

roughly constant flux, are confirmed by independent observations carried out by the NRAO group at 2,695 MHz (the Westerbork results were obtained at 1,415 MHz). The accumulation of data has also enabled Braes and Miley to obtain an improved estimate of the position of this radio source, which agrees well with those of the X-ray source and the star HDE 226868.

CANCER RESEARCH

Pre-malignancy

from a Correspondent

At the fourth meeting of the series on the principles of clinical and experimental oncology organized by the Institute of Cancer Research at the British Museum (Natural History) on January 31, Dr A. Spriggs (Churchill Hospital, Oxford) and Dr A. C. Allison (Clinical Research Centre, Northwick Park Hospital) were the clinically and experimentally orientated speakers who discussed pre-malignancy.

Dr Spriggs pointed out that latency in relation to tumour induction is a real phenomenon. He contrasted Wilms tumours of the kidney, which can be latent at birth and which can only have had a nine months induction period, with squamous cell carcinoma of the bronchus caused by smoking which has an average of forty years between the onset of smoking and the appearance of the tumour. Dr Spriggs concentrated his attention on carcinoma *in situ* of the cervix uteri. Investigation of this complex lesion is facilitated by its relative commonness. Various dysplasias can be recognized which can either be thought of as progressive steps in the development of the full-blown lesion or perhaps independent phenomena—the evidence is inconclusive. In carcinoma *in situ* there are variations in cell size, in patterns of mitotic activity, degree of lymphocytic infiltration and invasiveness in the forms of "micro-carcinoma". Aneuploidy is common and overall on histological and cytological criteria carcinoma *in situ* could be diagnosed as malignant.

Dr Spriggs, however, went on to point out that the clinical evidence is less decisive. Eradication by surgery is effective and permanent. Untreated lesions sometimes progress to invasive carcinoma, but there are many difficulties in being sure of the accuracy of the initial diagnosis in terms of the location of the original biopsy. The indications are that there is a variable latency in the emergence of fully malignant carcinoma but good statistics are hard to come by.

Dr Spriggs concluded his talk with an analysis of chromosome changes in carcinoma *in situ*. Again the evidence is inconclusive but it tends to show that clonal development does occur and that the same population of cells can be found in the anterior and posterior lip of the cervix in spite of general lack of evidence of invasiveness. Dr Spriggs felt that if his findings of widespread chromosomal abnormality in malignant or pre-malignant lesions are valid the possibilities for reversion are negligible. He likened the phenomenon to a pack of cards within which a particular

sequence existed—one shuffle disturbed the sequence and no amount of further shuffling was likely to restore the original state.

Dr Allison spoke first of minimal deviation hepatomas in experimental animals. Sometimes these are chromosomally normal, sometimes not. In the normal animal the response of the liver enzyme tyrosine transaminase to manipulation of the amount of protein in the diet always takes a particular form. Hepatomas are very variable in relation to the same property—some respond in the normal manner, others do not. There is no correlation between chromosomal abnormality and enzymatic response. Furthermore, it could not be said in non-responsive tumours that the gene(s) controlling the production of the enzyme has been deleted because injection of glucagon and cortisone lead to production of enzymes. Study of minimal deviation tumours leads to the conclusion that each tumour seems to have its own natural history—a point which was stressed later by Dr S. Brenner (MRC Laboratory of Molecular Biology, Cambridge), the chairman of the meeting.

Attempting to ascertain the clonal nature of tumours, Dr Allison spoke of studies with two isoenzymatic variants

of glucose 6-phosphate dehydrogenase, which could be classified as *A* or *B*. In human females, heterozygous for the sex-linked gene determining these enzymes, it has been shown that individual cells produce either *A* or *B* but not both. Furthermore, once the differentiations step leading to production of one or the other enzyme has been taken in a cell clone it does not seem easily reversible.

Dr Allison went on to describe studies of chronic myelogenous leukaemia in heterozygous females which showed that in any individual all the abnormal cells and the erythrocytes from the same patient had one but not both of the enzymatic phenotypes. The inference is that a causal agent affects a stem cell which gives rise to both myeloid and erythroid cells. One manifestation of the change is the Philadelphia chromosome characteristic of the myeloid and erythroid cells in such patients. On this basis chronic myelogenous leukaemia is a clonal disease. In cases of Burkitt's lymphoma cells are either *A* or *B* in a patient but not both, even when samples are taken from tumours from different sites—again the inference is that the tumours are monoclonal. Similar studies on trichoepithelioma and neurofibromatosis revealed the poly-

Genetics of Nervous Systems

THE structure and function of nervous systems in multicellular animals must, like any other trait, be under genetic control and therefore, in theory at least, it should be possible to dissect the genetics of nervous systems by exploiting appropriate mutants. The direct approach is to isolate sets of mutant strains of animals such as *Drosophila* and then study the behaviour of the mutants and of hybrids obtained by mating. Alternatively, one can exploit the techniques of somatic cell genetics and analyse the biochemical and physiological properties of hybrid cells obtained by fusing in pairs neural cells, which in culture retain at least some of the properties of nerve cells *in vivo*, or by fusing such cultivated nerve cells with cultivated fibroblasts and other cell types. As Minna, Glazer and Nirenberg report in next Wednesday's *Nature New Biology* (February 23) this approach holds great promise.

Minna *et al.* fused mouse neuroblastoma cells to mouse L cell fibroblasts, using Littlefield's HAT selection procedure to eliminate from the culture all but the hybrid cells. They then assayed the hybrids for enzymatic activities and other phenotypic characters contributed to the hybrids by the neuroblastoma cell parent. Furthermore, the chromosome constitution of each hybrid clone was recorded although

the karyotypes of mouse neuroblastoma cells and mouse L cells are not very different so that it is difficult to decide which chromosomes in the hybrids come from which parental cell type.

Minna *et al.* found that although the expression of many of the differentiated functions of neuroblastoma cells is extinguished by fusion with L cells three functions persist. The hybrid cells not only retain acetylcholinesterase activity but also may have electrically excitable membranes and may produce cytoplasmic processes containing neurites. Hybrid clones defective in one or more of these three functions were readily isolated, most having fewer chromosomes than hybrids exhibiting all three properties, and that means that it should be possible to discover which of these functions can be expressed independently of the others.

From the pattern of properties of their various hybrid lines Minna *et al.* are already able to propose a tentative sequence for the steps leading to the maturation of neuroblasts to neurones. And greatly encouraged by these results they suggest that it may be possible to produce hybrids of normal neurones and cells of established lines and perhaps eventually analyse *in vitro* the genetics of complex processes such as the formation of synapses and nerve networks.

clonal nature of these diseases. Dr Allison felt that understanding of internal regulatory mechanisms which are likely to affect the growth of some tumours in their early stages is inadequate, but suggested that the immune response is likely to be the most accessible to study.

REPRODUCTION

Monogamous Mosquito

from our Insect Physiology Correspondent

THE female of the yellow fever mosquito (*Aedes aegypti*) has often been described as mating repeatedly from very soon after emergence throughout a life which may last several months. Some forty matings before the first blood meal, fifty successful copulations with eleven different males during one hour, are typical records. But during the past few years a very different picture has emerged. In 1967, Craig (*Science*, **156**, 1499) found that a substance from the male accessory glands, received during mating, rendered the female permanently refractory to further insemination. This substance turned out to be a peptide to which the name "matrone" has been given. The effect can be produced in the absence of copulation by implanting the male glands into the abdomen of the female mosquito.

The nature of the refractory state has been studied in detail by Gwadz *et al.* (*Biol. Bull.*, **140**, 201; 1971, and *J. Insect Physiol.*, **18**, 152; 1972). It had been suggested that the female ejected the sperm received at all subsequent matings after the first; but it now seems that no sperm is received. The positioning of the vaginal orifice guarded by the cerci is such that the male cannot copulate successfully with the refractory female; the apparent repeated copulations of females are merely mechanical "couplings", of the same duration as copulation (about 12 seconds) but no sperm transfer takes place.

The female mosquito is indeed monogamous. On emergence from the pupa she is refractory to copulation; she becomes receptive within about 40 hours. This change results from the action of the juvenile hormone and can be speeded up to occur within 8 or 12 hours by treating the pupae or the freshly emerged adult females with a synthetic analogue of the juvenile hormone. Conversely, as was shown by Lea (*J. Insect Physiol.*, **14**, 305; 1968), the onset of receptivity can be prevented by removing the corpora allata from the newly emerged female and can be restored by re-implanting the glands. The peptide pheromone "matrone" from the male accessory glands exerts its action on the terminal

ganglion of the abdomen, which has long been known to control the mating behaviour of insects. It acts by the nervous route; and if the terminal ganglion is excised from a refractory female she will engage in repeated copulations so that the bursa copulatrix becomes distended with sperm received from several males. A similar cycle of controlled insemination was described in the housefly *Musca domestica* by Adams and Hintz (*J. Insect Physiol.*, **15**, 201; 1969).

It is remarkable that the refractory female continues to attract males which attempt copulation, whereas in other insects the refractory female is usually unattractive. Perhaps the reason is that in many insects mating is induced by male and female sexual scents, or pheromones, which are not secreted during the refractory state, whereas in mosquitoes the male is attracted by the flight tone of the female and this remains unchanged. It has long been known that male mosquitoes will endeavour to copulate with a tuning fork emitting the appropriate note.

ANIMAL LEARNING

Learning Laws Repealed

from our Experimental Psychology Correspondent

THE past two or three years have for many psychologists seen the abandon-

ment of some of the previous goals of learning theory. Partly, at least, animal learning had been studied in order to derive general laws of learning, applicable in all situations. Recent work by P. Rozin and J. Kalat of the University of Pennsylvania (*Psychol. Rev.*, **78**, 459; 1971), however, squarely rejects this approach. These authors point out that though sharing some common principles, learning mechanisms and their characteristics are for the most part various. Having been subject to selective pressures of widely different environments, these mechanisms, rather than exhibiting general laws of some slightly mysterious process called learning, represent specific adaptations of different animals to particular ecological problems.

Rozin and Kalat deal principally with how animals learn to avoid poisons and the related issue (and area of their experiments) of how they learn to select diets which relieve vitamin deficiencies. In this kind of learning some alimentary or other internal sensation such as a feeling of illness becomes associated with the taste or smell of a substance or substances most recently eaten. What is striking about the phenomenon is not just the efficiency with which it taxes the ingenuity of pest exterminators, but that the learning requires as little as one trial, spans delays of many hours between the occurrence of the taste and the onset of illness, and appropriately associates the internal sensation with

Putrescine, a Cell Growth Factor

MOST cells when placed in tissue culture require serum for their survival and proliferation and over the past several years numerous attempts, meeting with varied success, have been made to identify and to isolate those molecules or macromolecules which keep cells alive in culture and trigger cell division. Serum is, however, not only a complex but also a variable mixture of molecules and in order to identify cell growth factors cell biologists have turned their attention to so-called conditioned medium—medium which has been in contact with living cells. As Pohjanpelto and Raina, for example, describe in next Wednesday's *Nature New Biology* (February 23) medium conditioned by adult human skin fibroblasts contains a factor, presumably made and secreted by the cells, which stimulates the proliferation of embryonic human fibroblasts.

The maximal yield of this factor is apparently obtained when medium is conditioned for 5–7 days. The growth factor passes through dialysis tubing, and its behaviour on 'Sephadex G-15' columns confirms that it has a low

molecular weight. Furthermore, the factor is not particularly heat labile, neither is it soluble in ether or phenol but it can be extracted with alkaline butanol.

This last mentioned finding, together with the fact that polyamines have been shown to prolong the multiplication of Chinese hamster kidney cells in culture, led Pohjanpelto and Raina to think that the growth factor in their conditioned medium might be a polyamine. Pursuing this possibility they compared the effect of growth factor with the effect of various polyamines and found not only that putrescine partially mimics growth factor but also that the conditioned medium contains amounts of putrescine significantly in excess of the amount in unconditioned medium. They suggest, therefore, that the growth factor released from adult human skin fibroblasts is probably putrescine. They further suggest that it might be advantageous to add this polyamine as a supplement to culture media because its availability may determine the extent to which at least some sorts of cells can proliferate *in vitro*.

something eaten. Thus a rat will avoid tastes of foods or liquids it has consumed if illness follows within about 12 hours, but it will not avoid a visual or auditory stimulus following illness. This is not the same mechanism as that which copes with avoidance of noxious stimuli arising in the outside world (as opposed to within the gut), because a rat readily learns visual or auditory cues to avoid a place in which it has been shocked, but on the other hand the rat is not induced to refrain from eating food that has a particular taste by shocking it.

In learning which foods to eat to restore a vitamin deficiency, additional special mechanisms come into play: if offered a choice between four different diets, the rat, rather than taking some of everything, samples them one at a time, waiting several hours or days before trying each different food. Using this strategy the rat usually comes to select (and eat exclusively) the diet containing the needed vitamin.

This phenomenon of alimentary learning has characteristics which seem exactly fitted to its role in avoiding poisons and recovering from vitamin deficiencies. The ability to bridge long delays between the taste or smell and whatever alimentary sensations occur is appropriate when something one ate takes hours to have its ill effects. The association with the properties of the food itself rather than sights and sounds occurring at the time of eating is also entirely appropriate. Obvious though this may seem, neither of these findings fits into previously derived general laws of learning, from which one would conclude that long delays between conditioned and unconditioned stimuli would prevent learning altogether, and that any arbitrarily chosen stimulus (visual, auditory, gustatory and so forth) could control any arbitrarily chosen response such as avoidance of eating or drinking.

This, by now traditional, approach in psychology of regarding the Skinner box (or its equivalent) as a microcosm in which to discover universally applicable laws of learning, evidently needs at least to be supplemented by a more characteristically ethological approach of studying what an animal learns in the context of his particular natural environment. The idea that what can be learned easily is governed by the principle of biological appropriateness is already being incorporated into one of the other pervading traditions of learning theory, that of using principles drawn from animal research to tackle applied problems with people. Rozin and his colleagues (*Science*, **171**, 1264; 1971) have already claimed some success in bringing the idea to bear upon solving children's initial difficulties with reading, by having the children learn novel symbols representing whole words, rather than phonemes.

TRYPANOSOMES

A New Stage?

from a Correspondent

FOR the past 60 to 70 years there have been sporadic reports of the existence of "occult" stages (usually rounded amastigote forms) in the life history of the African sleeping sickness trypanosomes (subspecies of *Trypanosoma brucei*). Such accounts have usually been dismissed as referring only to so-called degenerate or involution forms, and it was generally believed that the only stages occurring in vertebrate hosts (including man) were the well-known trypomastigotes of the peripheral blood and tissue fluids.

Soltys and his colleagues, however, at the University of Guelph in Canada (*Trans. Roy. Soc. Trop. Med. Hyg.*, **63**, 490 and 495; 1969; and **64**, 692; 1970), have claimed recently to have isolated infective organisms of *T. brucei* from homogenates prepared from organs of infected rodents and filtered through membranes with pores too fine to pass the conventional trypomastigotes. Wéry and Kayembe (*ibid.*, **65**, 409; 1971) in Zaire (formerly Belgian Congo) briefly reported amastigotes and sphaeromastigotes (rounded forms with a flagellum) in the brains of chronically infected rats. Now Ormerod and Venkatesan, at the London School of

Hygiene and Tropical Medicine, have published a detailed account of similar forms in capillaries of the choroid plexus (the vascular roof of the third and fourth ventricles of the brain) of rats infected with strains of *T. b. rhodesiense* isolated from Botswana, and have proposed a new life cycle for this species to take account of these forms (*ibid.*, **65**, 722 and 736; 1971).

Briefly, Ormerod and Venkatesan found amastigotes and sphaeromastigotes in the choroid plexus of rats 48 and 64 hours after their inoculation with infected blood. These forms (illustrated by coloured photomicrographs) were not found at 72 or 88 hours, but were present again at and after the 96th hour. They were extracellular, and present in clumps associated with thrombi in the vessels. It is characteristic of the strains which Ormerod and Venkatesan studied to produce an initial "flush" of trypomastigotes into the peripheral blood between 72 and 96 hours after inoculation with bloodstream forms, followed at 144–168 hours by the disappearance of most of these trypomastigotes and then a relapse initiated by a second "flush" at about 192 hours. Ormerod and Venkatesan thus hypothesize that some of the inoculated trypomastigotes give rise by multiple division to rounded forms in the choroid plexus, while the remainder of the inoculated forms are destroyed; the rounded parasites then "unroll" into

Epigenetic Origin of BUdR Resistance?

THE suspicion that mutants induced in tissue culture cells *in vitro* are not all they might be has become increasingly widespread. The high frequency with which they are induced, their high reversion frequency and the continuing lack of direct evidence that such a mutation is actually in an appropriate structural gene have all conspired to spread an air of unease among somatic cell geneticists. This atmosphere can only be reinforced by a report by L. Mezger-Freed in next Wednesday's *Nature New Biology* (February 23).

Mezger-Freed shows that the bromodeoxyuridine resistant variants induced in the haploid frog lines previously described by Freed and Mezger-Freed are likely the result of permeability changes of non-mutational origin. Using nitrosoguanidine, ethylmethanesulphonate, and a variety of acridine half-mustards Mezger-Freed shows that there is no consistent difference between the frequency of resistant variants produced in the haploid lines compared with some genetically related pseudodiploid lines. Some haploids show rather more mutants, some rather less, but certainly nothing like the million-fold effect that might have been expected if bromo-

deoxyuridine resistance was being acquired by recessive mutations in the structural gene of the enzyme thymidine kinase.

This argument is also strongly confirmed by Mezger-Freed's measurements of thymidine kinase activity in the resistant variants which she finds on average to be about two-thirds the value of the parent line.

Mezger-Freed argues that the resistant variants result from an inheritable membrane change, and draws a parallel with puromycin resistant variants which she has also recently described (*J. Cell Biol.*, **51**, 742; 1971). There can be no doubt that the temptation is strong to compare these phenotypic shifts with the process of cellular differentiation; in this case too there are relatively stable shifts in gene expression without, so far as is known, any change in the corresponding structural genes.

It should be noted, however, that this report describes only bromodeoxyuridine resistance, and it is known that under certain conditions this drug has singularly unusual effects on the expression of a cell's differentiated functions (Silagi and Bruce, *Proc. US Nat. Acad. Sci.*, **66**, 72; 1970).

trypomastigotes and emerge into the peripheral blood to produce the 72–96 hour parasitaemia. During the 144–168 hour remission, a second “crop” of rounded forms develops in the choroid plexus to initiate similarly the relapse at 192 hours.

Ormerod and Venkatesan suggest that this cycle occurs only in strains of subspecies of *T. brucei* which produce relatively chronic infections, such as the West African forms (*T. b. gambiense*) and those from the southern part of the range of the East African form (*T. b. rhodesiense*); it probably does not occur in the virulent strains of *T. b. rhodesiense* which occur further north, nor in strains which have become “rodent-adapted” by prolonged maintenance in laboratory hosts. This might explain the failure of most previous investigators to observe it.

Ormerod and Venkatesan's evidence for the presence of these forms in the choroid plexus seems incontrovertible. But several questions remain to be answered about the proposed life cycle. First, the viability of the “occult” amastigotes and sphaeromastigotes is suggested so far only by their staining properties and the motility of the sphaeromastigotes. Second, it is difficult to understand why the trypomastigotes inoculated into the rat to infect it do not themselves immediately commence binary fission, as they do after the first “occult” cycle. Third, the work is based on infections initiated artificially by the inoculation of infected blood; what happens naturally when mammals are infected by tsetse flies? If these stages are truly integral to the life cycle, their discovery could clearly be of great importance in relation to chemotherapy and also to the organism's evasion of its host's immune response.

CHROMOSOMES

Going Macro

from a Correspondent

UNDER certain circumstances, one of the two genetically identical micronuclei of the ciliated protozoan *Stylonychia* changes into a macronucleus. The DNA content increases about fourteen-fold and results in the formation of polytene chromosomes which become transected into short lengths by membranous partitions between each of the chromomeres. Immediately after the breakup of the polytene chromosomes, 93 per cent of the DNA is destroyed and after a few hours the remaining 7 per cent of the macronuclear DNA replicates many times until finally the DNA content is increased to about sixty-five times the amount in the original micronucleus.

Prescott *et al.* (*Chromosoma*, **34**, 355; 1971) showed that although the micronuclear DNA remains as long pieces,

the macronuclear DNA is reduced to small pieces about 0.78 μ m long. This observation is quite consistent with the observed transection of the chromomeres, and the question arises as to the nature of the reduction and subsequent replication of the macronuclear DNA. Is the replication confined to only a selected portion of the DNA, or are all sequences equally amplified?

This question has now been clearly answered by Bostock and Prescott (*Proc. US Nat. Acad. Sci.*, **69**, 139; 1972), who have extracted the micronuclear and macronuclear DNAs and banded them in an analytical CsCl equilibrium gradient. The micronuclear DNA quite clearly has at least four different density components at 1.699, 1.701, 1.704 and 1.709 g/ml., whereas macronuclear DNA is an almost symmetrical band with a peak at 1.701 g/ml. Bostock and Prescott exclude the possibility that the extra DNA

bands seen in the micronuclear DNA preparations are mitochondrial DNA or DNA from food vacuoles in the *Stylonychia* cytoplasm, and they are careful to exclude the possibility of polysaccharide contaminants.

It seems therefore that the transformation of the micronucleus to the macronucleus has entailed massive amplification of a selected density species. Whether or not the new macronucleus lacks copies of the other micronuclear DNA density species, as the authors claim, is not quite so clear because their presence would have been obscured by the amplified copies of the 1.701 g/ml. species. The answer to this question will not be apparent until sufficient DNA can be extracted from the micronucleus for hybridization experiments. The fact remains that *Stylonychia* presents a very clear example of selective amplification, although the manner in which this occurs remains a mystery.

Separation of Cytotoxic Cells by Adherence

SEPARATION of populations of lymphocytes into two different piles is a useful exercise which is greatly facilitating the study of heterogeneity of lymphoid cell populations and also understanding of the cellular basis of immune responses. One of the first methods, devised by Wigzell, involved the passage of primed or non-primed lymphoid cells down antigen coated columns. The cells which emerged from the bottom of the column were shown to have been depleted of the capacity to respond to the antigen on the column. More recently, A. Basten, J. Sprent and J. F. A. P. Miller (*Nature New Biology*, **235**, 178; 1972) have shown that if lymphoid cells are incubated in serum containing antibody and subsequently passed down a column coated with the antigen for which the antibody is specific some lymphocytes are retained in the column. The method of Wigzell removes B cells which have the capacity to respond to a particular antigen; that of Basten *et al.* also removes B cells but probably all of them in a given cell population because B cells have the capacity to bind immunoglobulin. In next Wednesday's *Nature New Biology* (February 23), Clark and Kimura and Lonai, Wekerle and Feldman describe similar methods but ones in which the basis of separation is the capacity to respond to foreign cells *in vitro*.

If rat lymph node cells are plated out onto mouse fibroblast monolayers *in vitro*, it can be observed that a proportion of the plated cells becomes adherent to the monolayer within the first day. If the non-adherent cells are removed and plated onto a fresh monolayer of the same phenotype they can

be shown to develop less cytotoxic capacity than that demonstrated by the adherent cells. Although the capacity of the supernatant cells to respond to the specific target is reduced, however, these cells are still capable of responding against cells in an antigenically different monolayer. The ability of the adherent cells to respond to the monolayers of the sensitizing cells is amply demonstrated; what is not so clear is whether the adherent cells are restricted in their capacity to respond to a different target. Clark and Kimura's results suggest no such restriction and in fact hint at heightened capacity to respond—this could, however, be attributed to cross-reactions because of some antigens held in common by the different target layers.

The adherence itself commences in as little as 30 minutes after the initiation of the cultures. Various replating experiments are described in which supernatant cells were successively absorbed on monolayers, thus completely eliminating their capacity to respond to the specific target, and reducing their capacity to respond to any target.

The method described by these authors is broadly similar to that recently published by Sprent and Miller (*Nature New Biology*, **234**, 195; 1971) in a completely *in vivo* situation. Sprent and Miller showed that when parental thoracic duct lymphocytes were injected into irradiated F_1 recipients the cells that emerged in the thoracic duct were restricted in their capacity to respond, being able only to react against antigens similar to those encountered in their first F_1 recipient.

Recent Developments in Forensic Science

A. S. CURRY

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The system whereby the police forces in England and Wales are able to request the services of experienced forensic scientists at a moment's notice to assist them in their enquiries is now more than thirty-five years old. At present there are more than four hundred scientists and technicians engaged in this work at the Metropolitan Police Laboratory and at the Home Office forensic science laboratories.

IN 1966 the Home Office Central Research Establishment (HOCRE) was formed with the principal object of providing a centre for research in forensic science. The aim was to ensure that scientists in the field had the research, development and information services necessary to keep their particular professional skills at the forefront. The research now going on at HOCRE complements the *ad hoc* research projects which have been encouraged in the regional laboratories as well as the researches at the Metropolitan Police Laboratory. At its inception HOCRE was the first establishment in the world devoted solely to this kind of research.

At present the staff at HOCRE numbers thirty-seven of whom eighteen are science graduates. The establishment is located at the Atomic Energy Authority site at Aldermaston. To ensure that the research programme is well founded and takes account of current advances in science and technology, the director of HOCRE has available to him the advice of a committee of academic and industrial experts, each pre-eminent in his field.

The work of the operational forensic scientist ranges from comparative studies of materials that may be taken, often inadvertently, to or from the scene of a crime by the criminal to fundamental analysis of trace material for absolute identification. The first type of work includes, for example, the examination of glass fragments, paint chips, hairs, fibres, inflammable liquids, plastics, rubbers, explosives, dyes, blood, semen, pollen and wood; and, in the second category, perhaps the best example is the identification of sub-nanogram quantities of poison isolated in cases of suspicious death.

In the early days of this type of work a microscope and an emission spectrograph made up the *sine qua non* of the laboratory, but today all the modern instrumental tools are necessary. The regional laboratories are equipped with apparatus for undertaking the usual analytical techniques such as X-ray diffraction, ultraviolet and infrared spectroscopy and all forms of chromatography, but one of the first projects at HOCRE has been to assess the place of new instrumentation in forensic science and, in many cases, to provide a service facility for regional laboratories—especially where expensive instruments or techniques are concerned. The proximity of HOCRE to the Herald Reactor at Aldermaston has, thanks to the co-operation of the Atomic Weapons Research Establishment, enabled the service to set up a radiochemical section and to study in some depth the potential of neutron activation analysis in forensic science. The ability of this technique to measure the concentration of arsenic in a few hairs was dramatized by

the work on Napoleon's hair, but at HOCRE it has a more practical effect in that analyses have recently confirmed arsenical poisoning in two cases in which verdicts of murder were subsequently returned. The very great sensitivity of the technique allows a concentration gradient of arsenic to be measured from the root of the hair to the tip and so it can be decided whether a single dose of arsenic was administered or whether the victim had received several doses in the months before death. The hairs must be aligned individually before measuring and cutting them and this can now be done accurately because only fifty hairs are required in the neutron activation analysis technique.

The classical methods of analysis for arsenic involving digestion with a nitric/sulphuric mixture followed by reduction to arsine, which is measured colorimetrically, are so relatively insensitive that many thousands of hairs are required in each analysis. Nevertheless, it had been found that the work described by the Royal Commission on Arsenical Poisoning (1903) contains quantitative data that agree well with those obtained using the latest activation techniques.

In many crimes, fibres as well as hairs may be transferred between assailant and victim and the forensic scientist uses optical microscopy to a large extent in his comparisons. A 'Stereoscan' electron microscope is, however, now available and much work is being done at the West Midland Forensic Science Laboratory at Birmingham, where the instrument is housed, to assess the practical use of this powerful tool.

Trace element analysis of hairs and fibres has been studied through contracts with the Atomic Energy Authority using neutron activation analysis and with Bradford University using neutron activation analysis and mass spectrometry. It is clear that many problems remain to be solved before either is fully operational, but enough has already been done to allow the use of these sophisticated techniques on suitable occasions. In one recent murder enquiry dog hairs found on the deceased were identical microscopically with dog hairs on the clothing of a suspect. Comparison of these hairs by neutron activation analysis by HOCRE staff confirmed that they were very similar and also similar to only two other dogs in the village. In all, hair from more than 150 dogs was collected by the investigating officers.

Other radiochemical techniques in use or under development for use in post-mortem tissue analysis include insulin, using ^{125}I , and digoxin using ^3H . In these techniques antibodies are raised in animals to a drug-protein antigen. A measured small amount of antibody is then reacted with a radioactively labelled drug and a measure taken of the quantity of radioactivity removed from solution. If unlabelled drug from a tissue or blood extract is then mixed with the radioactive drug and the antibody reaction is repeated, the amount of radioactivity removed from solution is correspondingly less by an amount depending on the quantity of unlabelled drug competing for the antigenic sites. By using these radio-immune assay techniques very high sensitivities can be obtained.

In the field of inorganic analysis, mass spectrometry as well as neutron activation have been found to be most valuable. An AEI MS702, using photoplates, was purchased for HOCRE three years ago and the electrical detection accessory has just been installed. Work has concentrated on an investigation of the trace element composition of very small glass fragments and this has been done in conjunction with a contract with the AEA which has complemented the work using neutron activation analysis.

This and many other projects being undertaken at HOCRE are perhaps best looked at in the context of an investigation in which a murderer has broken into a house in the course of his crime and fragments of glass and paint are later found in or on the clothing of a suspect. It is necessary not only to compare these samples with others from the house but also to assess their value as evidence. It was found that there were no data available on the natural occurrence of glass and paint on clothing of people who have not been involved in crime. Consequently a random sample of one hundred suits was brushed down on receipt at a dry cleaner's and the contents of pockets and turn-ups examined for paint and glass fragments. Next, techniques for measuring the known discriminatory properties of glass (refractive index and density) were re-examined using the latest equipment. In the course of this study a way was found of reducing analytical time by a factor of ten while significantly increasing the precision as compared with current methods; the cost was only small and the necessary equipment was immediately installed by the Home Office in all the operational laboratories. These physical parameters of the glass found in the random study were then examined and a factor incorporated to allow for the known statistical correlation between them. At this stage it became apparent that the current view, that the occurrence of a sliver of modern production window glass on clothing had little value as evidence, was incorrect. Indeed, surveys of the physical properties of glass in the natural environment—including glass from nearly 1,000 broken windows at scenes of fires obtained in the course of an AWRE study, and from 184 scenes of crime in the City of Liverpool—showed that even for the most common window glass, there is only a likelihood of about 1 in 3,000 of a piece more than 1 mg in weight occurring by chance on clothing. For less common glass, especially from older houses, the evidential value can be very high indeed. The need to marry this study with the mass spectrometry project was obvious and it has now been shown that modern glass can be easily differentiated by its trace element composition into its site of production, and that even day-to-day variations between glass from the same furnace can be detected.

This study emphasizes that the forensic scientist is often involved in analytical comparison—the absolute concentrations of trace elements in the glass, for example, are of minor significance; the essential need is for comparison. Conversely, absolute measurements of refractive index must be made so that valid statistical studies can be undertaken; this has been one of the projects of a glass quality control committee chaired by the director of a regional laboratory.

A total of 3,558 paint fragments were found in or on the one hundred suits used in the survey. At first sight this is a very large number; but the ability of the eye to differentiate colour is well known and empirical colour tests, coupled with analyses of the organic matrix by a study of its solubility in organic solvents and use of Curie point pyrolysis followed by gas chromatography, allowed a complete sorting of the fragments to be made.

Work on other techniques to study the inorganic composition of paint fragments is now being pursued using the laser-arc emission procedure with the Carl Zeiss LMA 1. With this instrument single layers of paint can be analysed as the laser can be focused on a cross section of paint less than 100 μm thick. The significance of the presence of small flakes of paint on clothing can, like glass, clearly be considerable and, indeed, if several layers can be seen and analysed the value as evidence can be as high as that of a fingerprint; if the paint is from a car it should be possible for the make, model and year of manufacture to be confidently stated—and manufacturing variations, even in such a mass produced product, can be detected. In these two limited areas of paint and glass analyses, a considerable number of man hours obviously has to be spent in amassing analytical data before statistically viable conclusions can be drawn.

The toxicologist in forensic science used to be essentially an analytical chemist with a knowledge of medical biochemistry. The trend nowadays, however, is towards a much deeper understanding of the chemistry of death and towards studies of biochemical lesions and their relationship to particular drugs. Histochemical localization of drugs coupled with a study of the effects of drugs on enzyme systems may seem to be an almost academic pursuit but accurate knowledge of the precise cause of death is an essential requirement in any poison murder charge. In the field of transportation medicine, HOCRE has an attached unit from the RAF Institute of Pathology and Tropical Medicine, and research on the localization and distribution of carboxyhaemoglobin in erythrocytes, aimed at interpreting the role of in-flight fire or equipment malfunction in accident reconstruction, is of value to the forensic scientist because it may provide a means of differentiating an acute, perhaps murderous, gassing from an accidental slowly developing carbon monoxide intoxication.

The need for an increase in the sensitivity of detection has become more acute with the advent of the modern drugs problem; the active principles of cannabis and LSD have dosages of the order of a few hundred μg and so, when only a few ml. of blood or urine can be obtained from, say, a 70 kg suspect, the forensic toxicologist clearly needs very high grade methods for the isolation and identification of sub-nanogram quantities of drugs and uncharacterized metabolites.

One project at HOCRE concerns the development of automatic methods of analysis for drugs in urine. There is a need for a capability to handle hundreds of samples a day instead of the limited number that can be dealt with in manual systems.

Several aspects of forensic science can benefit from similar automatic machines—from the screening of urine samples mentioned above to studies on drug excretion and metabolism kinetics, and the statistical evaluation of drug involvement in transportation accidents. Systems already developed and in use at HOCRE include fully automated systems for barbiturate, amphetamine and morphine analyses of urine at a rate of twenty samples per hour with an ability to detect excretion patterns after ingestion of sub-therapeutic doses. The potential of such systems is very great and rates of 100 samples per hour have in fact been achieved; experiments which had to be done singly because of the time required for manual analysis can now be done in sufficient numbers to give statistically useful information. There have been some surprises about the time of excretion after ingestion; a single dose of butobarbitone, for example, reached its maximum peak excretion within 3 h but its concentration in the urine remained constant for nearly 100 h before tailing away. The variation in metabolite to unchanged drug ratios with variables such as urinary pH, liver function and degree of habituation are topics that can now be investigated because of the availability of rapid automatic analysis; even if such investigations were attempted in earlier days, many months of tedious manual work were required. Ultraviolet and visible spectrophotometers, as well as spectrofluorimeters, are used as the detectors in automated systems, and very high sensitivities can be obtained—for morphine, for example, less than 2 μg in 10 ml. of urine.

One of the characteristic features of forensic science is the variety of problems that can occur, and the uses to which organic mass spectrometry has been put provide a good example of this; recent cases have included the identification of volatile inflammables from lung tissue from victims of a fire tragedy, a study of the Curie point pyrolysis products of fibres and plastics to enable the original structure to be deduced and the identification of a deposit in a jug of milk as xanthene-9-carboxylic acid amide, leading to its recognition as an unexpected hydrolytic degradation product of a certain drug. This powerful analytical tool is indeed proving its worth in forensic science—the last case typifies how a difficult fundamental analytical problem may come the way of the operational scientist. It is perhaps only a little premature to

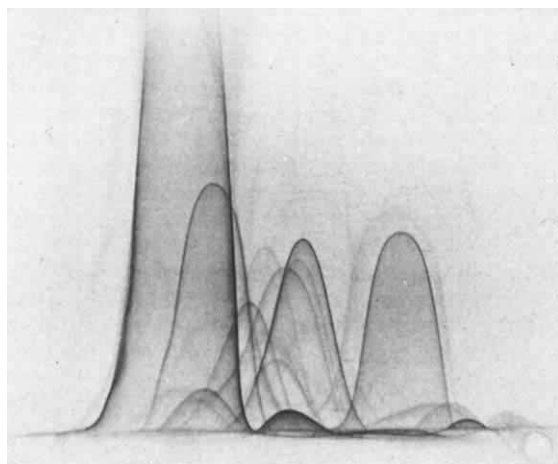


Fig. 1 The separation of proteins in human blood by electrophoresis in two dimensions.

look forward to the day when the forensic scientist will have a computerized mass spectrometer service at his fingertips in his own laboratory; in the case of England and Wales, such a service is already available at Aldermaston.

Formal distinctions between scientific disciplines are considerably blurred in forensic science. The electron microscope, for example, provides a means of studying the botanical features of the *Cannabis* plant at very high magnification and it may be possible to find structural characteristics that can help in detecting its source or some common identity with a control sample. The same samples may, however, also be subjected to a comparative gas chromatographic analysis giving quantitative measurements of the amounts of cannabinol, cannabidiol and tetrahydrocannabinol present. Again, atomic absorption analysis of metal swarf may be combined with mass spectrometry for trace element analysis as well as examination by the visible or electron microscope—the scientist looking for coincidence of striation marks that may enable him to say that the swarf came from a machine in a particular factory. That the swarf adheres to a murder weapon or is in the boot of a car suspected of being involved in an attack at the factory provides added interest. It can also mean that the time expended on the analysis becomes an important factor. Indeed, one of the challenges of the not too distant future will be that of organizing day-to-day availability of the major routine analytical tools suitable for use in the nanogram region (both in inorganic and organic analysis) for the regional forensic science laboratories. Development projects at HOCRE have clearly shown the advantages of mass spectrometry in both areas but at the same time have emphasized the well known fact that full time experienced teams of scientists with a continuous work load are required to maintain good results. Large industrial firms are able to centralize their expensive analytical instruments; the forensic science service may do so but it has also to take into account the legal rules of evidence that can require the personal appearance in court of the equipment operator or his supervising scientist.

Crimes of violence have recently been increasing and this means that more and more exhibits of blood, semen, hair and other biological materials are brought into the forensic laboratory by police "scene-of-crime" officers. In the laboratory the work load has also increased considerably with the introduction of more advanced serological and immunological techniques enabling many blood group and genetically determined enzyme systems to be studied on dried blood or semen stains. At the Metropolitan Police Laboratory, Miss M. Pereira has recently begun a study of advanced blood grouping tech-

niques for dried stains—with extensive emphasis on rhesus groups. At HOCRE emphasis has been placed on a study of blood proteins as a means of discriminating between the blood of individuals. The Laurell technique of crossed antigen-antibody electrophoresis separates the proteins present in 1 to 2 μ l. of blood and it has been shown that many of these are relatively stable in dry stains. This stability is an important feature in forensic science work because the suspect may only be connected with the victim of an assault through a few small blood stains on his jacket. In the Laurell method the results are obtained as a series of near Gaussian curves (Fig. 1) and computer analysis of these, now being studied under contract at the Cranfield Computing Centre, should enable quantitative blood protein analysis to become a feature of future investigations. The genetic nature of several protein systems also offers hope of an additional independent method of distinguishing between blood from different people. The existing techniques of blood grouping and enzyme measurement can occasionally give a discriminatory power of the order of 1 in 1,000. This is of great assistance to the investigating officer and sometimes makes it worthwhile to take blood or saliva from everyone in a small community in an effort to pick out the few individuals who, from their genetic makeup, fall within the suspect group.

It is still difficult, however, to determine from a blood stain whether the blood came from a male or female person. Protein analysis may eventually help—it is known, for example, that caeruloplasmin levels rise in women on the pill—but more recent work at HOCRE on visual marking of the Y chromosome using fluorescence microscopy shows promise. White cells are stained with quinacrine mustard and the Y chromosome turns out to be more deeply stained than the rest of the cell. A bright yellow fluorescent spot therefore reveals that the blood came from a male person. The use of drumsticks is also being studied so that positive indications may be obtained for both sexes. In a recent "blind" trial at HOCRE three *XY* persons were correctly placed in a search of twenty-four unknowns. Clearly a lot of work remains to be done because decomposition occurs when a blood stain ages and the failure rate soon becomes significant. It may seem at this stage that, as far as biological investigations are concerned, overdue emphasis has been placed on serious crimes of violence; but the spin-off is much wider—an anonymous letter writer, for example, needs to keep in mind that it may be possible to find the sex of the perpetrator by finding Y chromosomes and other characteristics in the cells among the saliva used to seal the envelope.

The forensic scientist has an interest in so many and such diverse areas of science that a centralized library and information service was quickly recognized to be one of the first requirements on the setting up of HOCRE, and a central collection of data and of physical samples of materials such as drugs was set up. More than 1,000 journals are regularly searched and extensive use is made of microfilm and microfiche in all laboratories; a computer based retrieval system especially tailored for forensic science is in the final stages of development. Clearly communication is very important—a well informed scientist is a desirable attribute in a court and knowledge that he is well informed provides the confidence that goes to the making of a good witness. With this and the needs of training in mind, video tape recording is now being used extensively in police forces and forensic science laboratories and they cooperate closely in making tapes for use in the training of police officers. A forensic scientist only works on the material that arrives on his bench and all his expert knowledge would be useless if the police officer did not correctly appreciate what material to send to the laboratory for examination.

The enormous growth in the case load of the forensic science laboratories during the past thirty-five years has shown how very important the service is in the task of preserving law and order; the forensic research and development programme at HOCRE aims to increase its effectiveness.

Photochemical Ozone and Sulphuric Acid Aerosol Formation in the Atmosphere over Southern England

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Photochemical air pollution of the kind which occurs in some urban environments in the United States has been observed in Britain during July 1971. The mechanisms involved may constitute an important part of the processes for the oxidation of sulphur dioxide in the atmosphere.

MEASUREMENT of oxidant in the atmosphere has been used for some years in the United States as an indicator of the occurrence of photochemical air pollution in urban environments. Levels of oxidant (chiefly ozone) in excess of 10 parts per hundred million (p.p.h.m.) are considered to be evidence of photochemical smog formation¹. By comparison, clean air ozone levels vary from 2 to 5 p.p.h.m. during daytime, falling to near zero at night².

It has generally been held that photochemical air pollution is unlikely to be encountered in Western Europe; because there is less sunlight and air temperatures are lower at these latitudes, smaller amounts of pollutant precursors (oxides of nitrogen and hydrocarbons) are emitted from motor vehicles. But some measurements made in Vlaardingen (in the Netherlands) have shown oxidant levels in excess of 10 p.p.h.m. on a number of occasions during 1968 when conditions were favourable for photochemical air pollution³. Further measurements made in 1969 clearly point to local photochemical production of oxidant in the atmosphere over the Western Netherlands⁴. No other systematic measurements of this type appear to have been made in Western Europe or the United Kingdom in recent years although a few observations of high ozone levels were made in Frankfurt/Main during the summer of 1967⁵.

Cox and Penkett⁶ have shown in laboratory experiments that when the photochemical smog reaction, involving unsaturated hydrocarbons and oxides of nitrogen, takes place in the presence of sulphur dioxide, oxidation of the sulphur dioxide to sulphuric acid occurs at a significant rate even with concentrations of hydrocarbons and oxides of nitrogen typical of the levels (about 0.1 p.p.m.) occurring in only moderately polluted atmospheres. A similar oxidation occurs in the presence of the ozone-olefin reaction⁷ which takes place in the dark. We present here evidence that (1) measurable levels of photochemically produced ozone occurred at ground level over Southern England on certain days during July 1971, and (2) at the same time the concentrations of oxidized sulphur dioxide in the form of sulphuric acid and sulphates were much higher than normal.

Measurements of ozone concentration were made with a chemiluminescent detector constructed at AERE Harwell to a design similar to that of Warren and Babcock⁸. Atmospheric air is drawn at 1 l./min into a glass reaction cell into which a stream of ethylene at 10 cm³ min⁻¹ is injected. A rapid chemiluminescent reaction occurs between the ethylene and any ozone in the air stream and the light emitted is measured

by an EMI type 9635 QD photomultiplier placed in optical contact with the cell inside a light-tight box. The current from the photomultiplier is amplified and displayed on a millivolt recorder. The instrument was calibrated against the amount of iodine liberated from a neutral potassium iodide solution using pure air-ozone mixtures and the response was found to be linear over the range 0–2 p.p.m. ozone after subtraction of the photomultiplier dark current. The latter varies somewhat with ambient temperature and is equivalent to about 3 p.p.h.m. of ozone. Provision is therefore made to measure the dark current by automatically stopping the air flow for a few minutes every hour. The instrument records from 0–10 p.p.h.m. ozone on its most sensitive range. It was

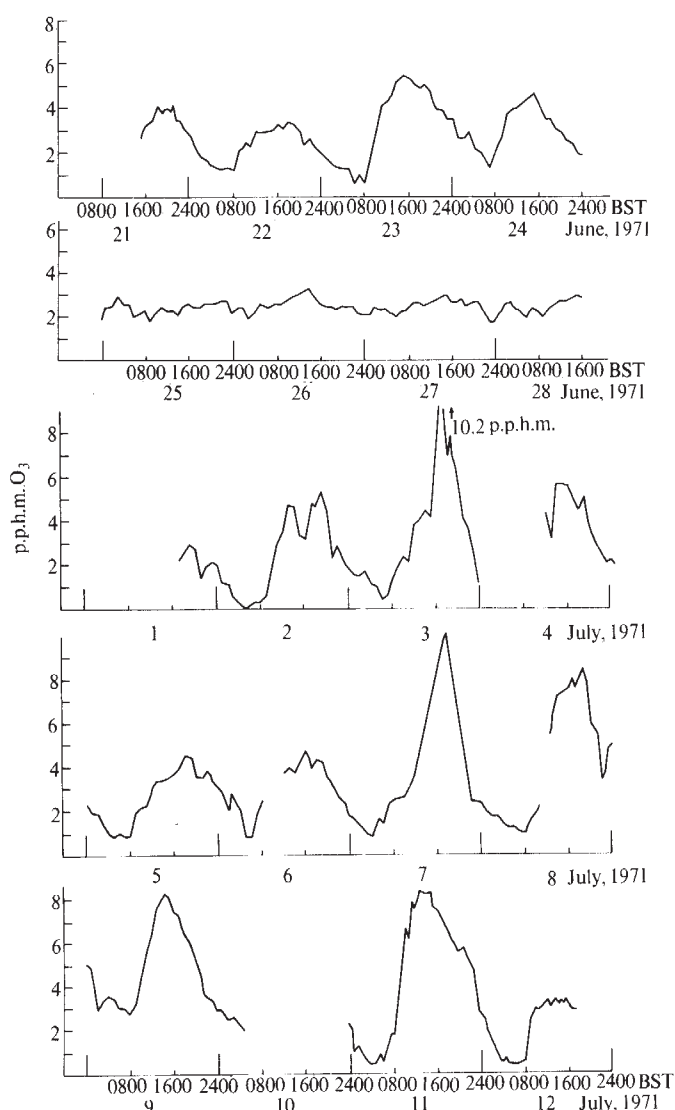


Fig. 1 Continuous daily ozone measurements.

set up in a first floor laboratory at AERE Harwell and sampled outside air 4 m from the ground through a 2 m length of 3 mm diameter nylon tubing. Preliminary tests showed that losses of ozone in this tube were negligible. A chemiluminescent method has the advantages that it is specific for ozone, has a high speed of response and operates unattended for long periods.

A continuous daily record of several meteorological parameters is made at Harwell in connexion with an air sampling programme. The latter includes, among other measurements, hourly filter samples of the atmospheric aerosol. The sampling site for these measurements is situated on grassland more than 300 yards from any local sources of pollution, roughly 1 mile from the A34 trunk road passing the entrance to AERE and roughly half a mile from the site of the ozone measurements. The filters for periods of interest were analysed for sulphate colorimetrically by reaction with a barium-thoronol complex⁹ and for total acid by direct titration with sodium tetraborate to pH 7 (ref. 10). The acid is assumed to be sulphuric acid because other common acids such as nitric or hydrochloric acids will escape collection on the filter paper at the concentrations observed. The contributions from motor vehicles of oxides of nitrogen and hydrocarbons were estimated indirectly by measurement of lead on the hourly filter samples, the lead being determined by X-ray fluorescence.

The concentration of radon in the atmosphere is also monitored hourly by measuring the radon daughter product activity on the air filters with a Geiger counter immediately after collection. Radon is emitted from the ground at a roughly constant rate and its concentration in the air gives an estimate of the rate of vertical mixing, high radon concentrations being associated with poor mixing and vice versa.

Fig. 1 shows the diurnal variation of hourly average ozone concentration during the period June 21–July 13, 1971. The record is not complete because of occasional failure of the pump to maintain a constant flow overnight. Also the instrument was disconnected from time to time for calibration purposes and for laboratory measurements. Sufficient information has been gathered, however, to draw some conclusions regarding ozone levels, their variation and dependence on meteorological conditions.

During the period of operation the hourly values varied from zero to maxima of 10 p.p.h.m. Minima were normally recorded during the early hours of the morning and maxima during the afternoon. These times correspond roughly to the daily maxima and minima in the extent of vertical mixing in the lower troposphere.

In Fig. 2 ozone and radon concentrations together with

various meteorological parameters are shown for two consecutive Saturdays, one June 26 when little photochemical activity occurred and the other July 3 when the ozone concentration reached 10 p.p.h.m., we believe as a result of photochemical ozone production. In Fig. 3 the corresponding synoptic charts for midday on the dates in question are shown together with the radio-sonde ascents from Crawley. June 26 was characterized by a fresh south-west wind and periods of rain with occasional sunshine. The radio-sonde ascent at midday showed an adiabatic lapse rate up to about 1,000 m at 1200 BST and by 1300 BST, when the surface maximum temperature was reached at Harwell, mixing extended to 1,500 m. Mixing was sufficiently good to reduce the surface radon concentration to 50 (arbitrary units) in the middle of the day and ozone concentrations remained between 2 and 3 p.p.h.m. throughout the 24 h period. On July 3 there was a light easterly wind with hazy sunshine in the morning and mixing was limited to a height of 700–800 m. Strong sunshine began at 1400 BST and the ozone concentration started to rise rapidly about 1 h later, reaching 10.2 p.p.h.m. at 1700 BST. The radon fell to a minimum value of 150 (arbitrary units) at the same time and, as shown later, concentrations of several pollutants were much higher than on June 26. Mixing on July 3 was evidently poorer and the higher ozone concentration observed on that day was therefore attributed to production near the ground rather than greater downward transport of stratospheric ozone.

Ozone in ground level air may be either natural ozone of stratospheric origin which has diffused down through the troposphere or it may be formed photochemically at low levels. Some measure of the stratospheric contribution can be obtained from the concentrations observed between June 25 and 28. During this period high wind speeds or the presence of cloud cover were sufficient to prevent the formation of nocturnal inversions, as shown by the temperature or radon records (see, for example, data for June 26 shown in Fig. 2) and mixing remained good during both day and night. Pollutant concentrations were low in a brisk south-west air stream (Fig. 4) and little photochemical activity occurred, as shown by the absence of any peaks in the middle of the day. The ozone concentrations observed during this period averaged about 2.5 p.p.h.m. It is interesting that the concentration returned to about this level between July 24 and 26 when similar wind and cloud conditions were again observed. It seems likely that this value represents a general level for the stratospheric component in the lower troposphere between late June and the end of July 1971 and any higher level must be due to photochemical activity.

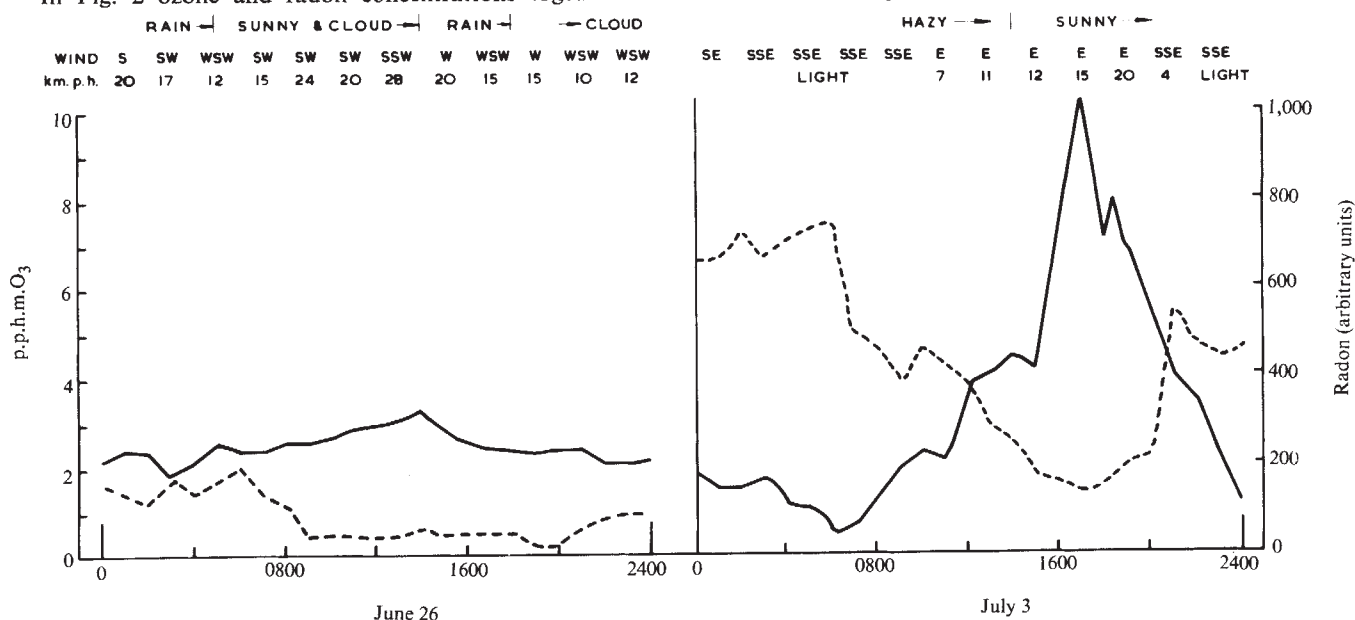


Fig. 2 Variation of ozone (—) and radon (---) levels.

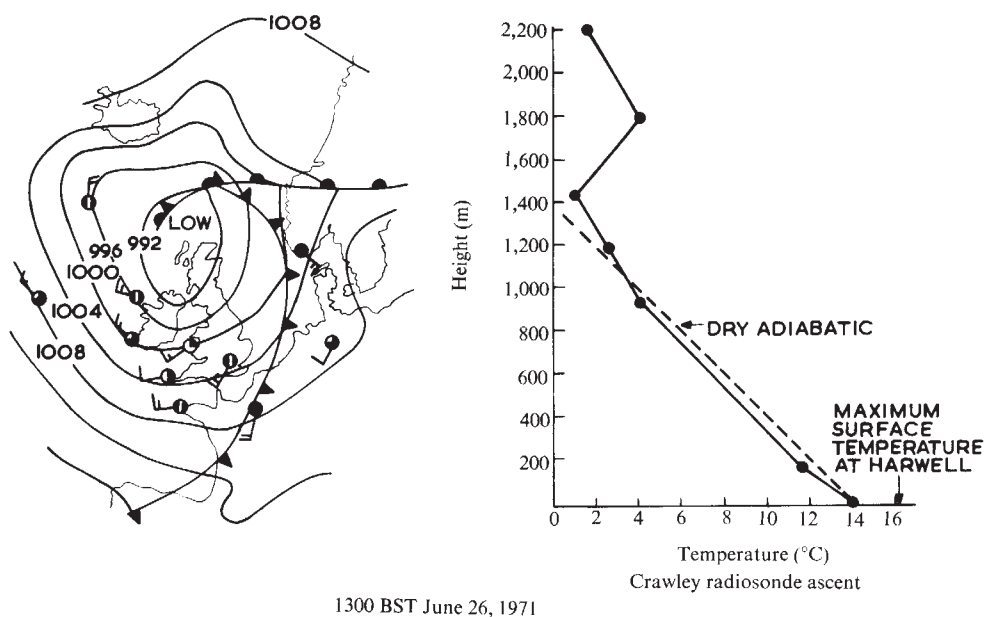
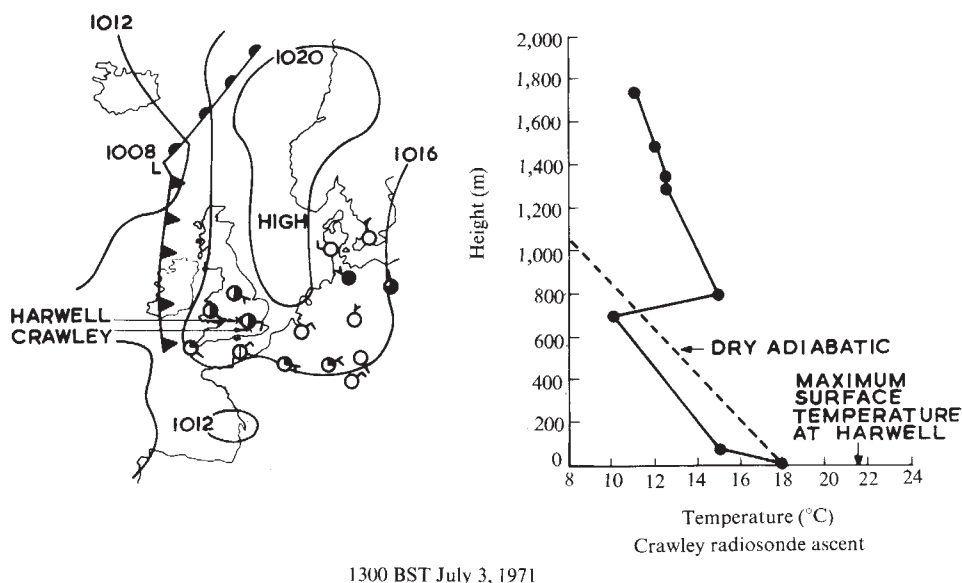


Fig. 3 Meteorological data for June 26 and July 3, 1971.



1300 BST July 3, 1971

Table 1 shows the frequency distribution of the maximum hourly average ozone concentrations on 35 days between June 21 and August 17. The maximum hourly value exceeded the background level on about half of the days and exceeded 8 p.p.h.m. on 6 days, on 2 of which (July 3 and July 7) values in excess of 10 p.p.h.m. were recorded. All these days were characterized by bright sunlight and light winds, that is conditions favourable for the occurrence of photochemical activity. According to Junge² maximum hourly ground level ozone concentrations in clean air do not normally exceed 5 p.p.h.m. and values above 7.5 p.p.h.m. are extremely rare. We conclude therefore that measurable amounts of photochemically produced ozone were present at Harwell during this period. For comparison, the hourly average concentration of oxidants (which is close to that for ozone) exceeded 10 p.p.h.m. in Los Angeles on 48.5% of days during 1964–1967 and reached a maximum value¹ of 58 p.p.h.m. The recommended maximum allowable 1 h average concentration for the United States,

not to be exceeded more than once per year¹¹, is 8 p.p.h.m. This level was reached or exceeded at Harwell on 6 days during the 35 day period of measurement. Levels are likely to be still higher nearer to major sources of vehicular pollutants, for example, the London conurbation, especially on days when atmospheric mixing is worse than encountered in this study.

Figs. 4, 5 and 6 show ozone and atmospheric aerosol data for 3 days, June 26 and July 3 and 7. On June 26 (Fig. 4) in the strong westerly air stream, the concentration of SO_4^{2-} was

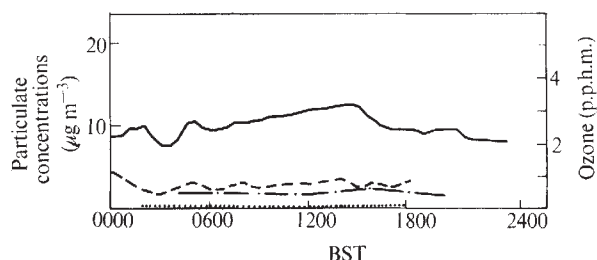


Fig. 4 Chemical data for June 26, 1971. —, O₃; ---, SO₄²⁻ (total); — · —, Pb (× 10); . . . , H₂SO₄.

Table 1 Frequency Distribution of Maximum Ozone Concentrations

Max O ₃ (p.p.h.m.)	0–1.9	2–3.9	4–5.9	6–7.9	8–9.9	10–11.9
No. of days	1	14	13	1	4	2

low ($3 \mu\text{g}/\text{m}^3$) and approximately constant. The lead and sulphuric acid were at or below the limits of detection.

On July 3 (Fig. 5) the aerosol levels were much higher than on June 26 and rather variable. Total sulphate reached $50 \mu\text{g}/\text{m}^3$ near midday, fell somewhat around 1600 BST and increased thereafter reaching nearly $70 \mu\text{g}/\text{m}^3$ by midnight. Hydrogen ion concentration, expressed as sulphuric acid, showed a much closer correspondence in behaviour with that of ozone, falling off like the latter after reaching a maximum at 1700 BST. Lead levels were higher than on June 26 reaching a maximum of about $0.85 \mu\text{g}/\text{m}^3$ at 600 BST and another of $0.55 \mu\text{g}/\text{m}^3$ at 1600 BST. The relative humidity fell to a minimum of 53% at 1700 BST and remained below 80% from 0900 BST to 2100 BST.

Fig. 6 shows that the high ozone levels on July 7 were also accompanied by an increase in sulphate and sulphuric acid although the aerosol levels were considerably lower than on July 3. The sulphuric acid showed a well defined maximum which corresponded with the maximum ozone level at 1700 BST. Lead concentrations which were measured by a more sensitive analytical technique than for the previous days reached a maximum of $0.75 \mu\text{g}/\text{m}^3$ at 1800 BST. Relative humidities were below 85% from 1000 to 2400 BST and 60% from 1400 to 2100 BST.

Statistical treatment of the data revealed a correlation coefficient of 0.71 ($P < 0.01$) between ozone and sulphuric acid concentrations for the days of July 3 and 7. The simultaneous decrease of ozone and sulphuric acid concentrations indicates that each have similar atmospheric residence times of a few hours. The ozone is destroyed by contact with the ground or by reaction with other pollutants such as nitric oxide while the sulphuric acid is converted to ammonium sulphate by reaction with ammonia released at or near the ground. Thus on July 3, for example, the decrease in the hydrogen ion concentration after reaching the maximum value was accompanied by a corresponding increase in the ammonium ion concentration necessary to maintain cation-anion balance.

The correlation coefficient for ozone against sulphate was only 0.31 and hardly significant. This is not unexpected as sulphate is removed from the atmosphere almost solely by rainfall and has a much longer mean residence time of several days at least¹². Sulphate concentrations are thus indicative of the history of an air mass for several days previously and advection of air in which oxidation of sulphur dioxide occurred

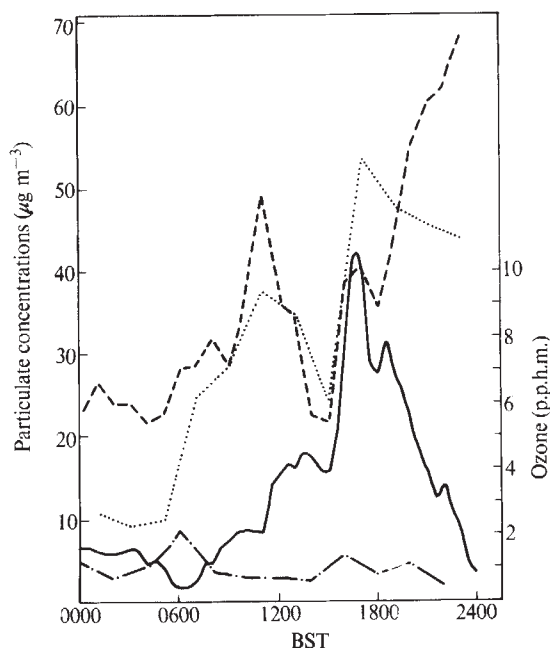


Fig. 5 Chemical data for July 3, 1971. —, O_3 ; ---, SO_4^{2-} (total); — · — ·, $\text{Pb} (\times 10)$; ..., $\text{H}_2\text{SO}_4 (\times 2)$.

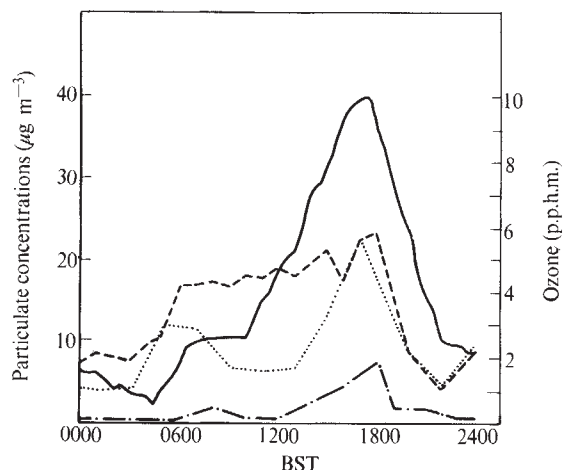


Fig. 6 Chemical data for July 7, 1971. —, O_3 ; ---, SO_4^{2-} (total); — · — ·, $\text{Pb} (\times 10)$; ..., $\text{H}_2\text{SO}_4 (\times 2)$.

at some distance complicates the pattern observed at a single station such as Harwell. Much of the sulphate on July 3, for instance, probably originated over Northern France and the Low Countries.

From the measured lead concentrations some estimate may be made of hydrocarbon and oxides of nitrogen concentrations before the start of photochemical activity on the assumption, which is not necessarily true, that motor vehicle emissions are the major source of all these pollutants in the atmosphere. A typical British car emits about 8 g/mile of hydrocarbons (measured as hexane), 2.5 g/mile of oxides of nitrogen expressed as NO_2 , and burns fuel containing 0.08 g of lead per mile (D. R. Fussell, personal communication). About 20% of the lead consumed is emitted as particles small enough to remain airborne for a significant length of time¹³. The maximum lead level of $0.75 \mu\text{g}/\text{m}^3$ on July 7 thus corresponds with $375 \mu\text{g}/\text{m}^3$ of total hydrocarbons and $117 \mu\text{g}/\text{m}^3$ or 6.1 p.p.h.m. NO_2 . The hydrocarbon figure, which excludes background methane, is equivalent to 63 p.p.h.m. v/v expressed as carbon or 6.3 p.p.h.m. as photochemically active hydrocarbons on the assumption that half the hydrocarbons are reactive¹⁴ and the mean carbon number in vehicle exhaust is five. According to Robinson and Robbins¹³, petrol driven engines under US conditions account for about half of total emissions of reactive hydrocarbons and a somewhat smaller proportion of oxides of nitrogen. Hence the actual values may have been a factor 2 higher or about 125 p.p.h.m. carbon and 12 p.p.h.m. NO_2 respectively. For comparison, on a similar July day in Philadelphia¹⁵ when levels of hydrocarbons, expressed as carbon, and nitrogen oxides reached about 200 p.p.h.m. and 16 p.p.h.m. respectively, the total oxidant reached 14 p.p.h.m. compared with 10 p.p.h.m. of ozone at Harwell on July 7. Taking into account the approximate nature of the calculation, these figures serve to show that pollutant concentrations were of the right order of magnitude to account for photochemical formation of the ozone observed on July 7. Better comparison awaits further experimental measurements.

Cox and Penkett⁶ found in their laboratory experiments that photochemical oxidation of sulphur dioxide occurred at a rate equivalent to 10% per hour in bright sunlight with initial concentrations of 10 p.p.h.m. and 3 p.p.h.m. of *cis*-2-pentene and nitric oxide respectively. Oxidation rates of a similar order may therefore be expected for the conditions of July 7. Non-photochemical modes of oxidation, particularly in mist and fog droplets, are no doubt important in the atmosphere in suitable conditions but the low relative humidities observed preclude such mechanisms on July 3 and 7. We therefore conclude that oxidation on these and other similar days was substantially due to the photochemical mechanism involving hydrocarbons and oxides of nitrogen, and that this constitutes an important general mechanism for the oxidation of sulphur dioxide in the atmosphere.

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Logarithmic Relationship of DDE Residues to Eggshell Thinning

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A concentration-effect relationship seems to exist between DDE in eggs and shell thinning. It seems to follow a mathematically similar pattern in different species though it seems to operate at different levels.

SHELL thinning has been recorded in eggs of the brown pelican (*Pelecanus occidentalis*) which were collected in widely separated nesting colonies¹⁻³, and it has been reported among numerous other species of wild birds⁴⁻⁹. The condition has been used as an indicator of population trend; thinning of 15 to 20% has been associated with declining populations of several species⁹.

A cause-and-effect relationship has been established between DDE (1,1-dichloro-2,2-bis (p-chlorophenyl) ethylene) in the diet and shell thinning; the associated residues in eggs of American kestrels (*Falco sparverius*) fed DDE were comparable with those in eggs of peregrines (*Falco peregrinus*)¹⁰ collected in the field. It is necessary to understand the quantitative relationships between residue content and shell thinning in order to evaluate the problem properly. We have studied this relationship as derived from a study of eggs of the brown pelican and we compare here these results with those reported for other species.

The questions to be explored are (1) whether a concentration-response relationship, paralleling the traditional dose-response relationship, in fact exists between DDE in eggs and the thinning of the shells, and (2) the nature of such a mathematical relationship. The amount of DDE in the egg is taken as an index to the concentration of residues in the female¹¹⁻¹³, the physiological processes of which determine shell thickness. DDE is the only residue considered here, because this residue consistently accounts for a significant percentage of eggshell thinning in these pelicans (our unpublished work).

Eighty eggs of brown pelicans were used in the primary analysis of the problem. Seventy eggs were collected in 1969 from twelve colonies, one in California, two in South Carolina and nine in Florida. Ten more eggs were collected from one of the South Carolina colonies in 1970. Analytical procedures for preparation, clean up, and residue analysis followed those

used at the Patuxent Wildlife Research Center¹⁴⁻¹⁶. The samples were prepared and analysed by Soxhlet extraction and clean up by acetonitrile partitioning and 'Florisil' column. The residues were separated and removed in four fractions from a silica gel thin layer plate. Sample fractions were analysed by electron capture chromatography on a column of 3% OV-1 on 'Chromosorb W' H.P., or on a column of 3% OV-17 on 'Gas Chrom Q'. The DDT metabolites were confirmed on a column of 3% XE-60 on 'Gas Chrom Q'. Some of the samples were treated with alkali to confirm DDT (1,1,1-trichloro-2,2-bis (p-chlorophenyl) ethane). Each of the three glass columns was 1.8 m long and the outside diameter was 0.64 cm.

The average recovery of the chlorinated insecticides and their metabolites from fortified eagle tissue ranged from 75 to 112%; the recovery value for DDE was 95%⁴. The pelican egg residues were not corrected for recovery. Residues are expressed on a fresh wet weight basis. We found that certain external egg measurements were significantly correlated with the weight of the contents of fresh eggs. The resulting regression equation was used to convert weight of the contents of addled eggs to a fresh wet weight basis (unpublished work of L. F. Stickel, S. N. Wiemeyer and L. J. B.). The statistical analyses were performed after the residues were transformed to logarithms.

The percentage of eggshell thinning was measured by comparison with the pre-1947 mean thickness as computed by Anderson and Hickey¹⁷. These means were 0.557 mm for Florida and South Carolina eggs and 0.579 mm for California eggs. Eggshell thinning occurred in eleven of the twelve colonies where eggs were collected. In eggs from the nine Florida colonies the mean change in thickness varied from a 13% decrease to a 0.4% increase. There was a 17% decrease in shell thickness of eggs from the two South Carolina colonies, and a 35% decrease in shell thickness of eggs from the California colony.

The thinning of eggshells of the brown pelican proved to be related to the concentrations of DDE in the eggs (Fig. 1). The relationship was essentially linear ($P < 0.01$) on a logarithmic residue scale. We used the following regression equation to describe this relationship:

$$\hat{Y} = b_0 - b \log_{10} X$$

where $\hat{Y}$ = % of pre-1947 eggshell thickness; b_0 = expected % of pre-1947 thickness at 1 p.p.m. of DDE; b = the regression

coefficient; and $\log_{10} X =$ p.p.m. of DDE on a fresh wet weight basis.

The significance of the regression indicates that the shell thickness decreases in a predictable manner as the DDE concentration increases. It also means that the percentage change was greater per unit of DDE when the concentration of DDE was lower. In other words, the lower concentrations were more effective. For example, the calculated percentage of thinning per p.p.m. of DDE is 4.2 at 1 p.p.m., 3.0 at 5 p.p.m., 1.9 at 10 p.p.m., and 0.4 at 100 p.p.m.

Analysis of covariance revealed no significant differences ($P > 0.05$) among slopes of the three regression lines that were plotted using data from each of the three states from which the eggs were collected. Thus, most of the variation in eggshell thickness (64%; $P < 0.01$) may be explained by the common regression used here.

The prediction of a no-effect level is also theoretically possible, but should be approached with caution because of the scarcity of eggshell measurements in eggs with lower residues, and the hazard of making predictions of X far removed from the mean¹⁸. The estimated no-effect level is 0.5 p.p.m., but the validity of this estimate is questionable because of the forementioned reasons. In the eggs from Florida that contained 0.52 p.p.m. of DDE, the thickness (0.64 mm) was slightly less than 0.65 mm, the maximum pre-1947 measurement; thus, the possibility of thinning was not ruled out completely in any of the eighty eggs. Anderson *et al.*⁴ indicated an apparent absence of a no-effect level of DDE on the eggshell thickness of the double-crested cormorant (*Phalacrocorax auritus*) and the white pelican (*Pelecanus erythrorhynchos*). The relationship of the logarithm of DDE to eggshell thinning also seems to apply to the prairie falcon (*Falco mexicanus*)⁵ and the double-crested cormorant⁴.

The relationship between log dose and response is often encountered and is in accord with toxicological and pharmacological theory¹⁹⁻²². The association between residue and response is poorly known.

The relationship between shell thinning and DDE residues in the eggs is particularly important in understanding the population status of the brown pelican.

The brown pelican population in South Carolina has been declining for at least 12 yr^{1,23} and the number of young fledged per breeding female in 1969 and 1970 was well below the estimated number necessary to maintain a stable population (unpublished work of C. Henry). The brown pelican in California experienced reproductive failure in 1969²⁻³, but the Florida population seems stable over the past 3 yr²⁴⁻²⁵.

The brown pelican seems to be unusually susceptible to shell thinning, a 15% thinning being associated with DDE residues between 4 and 5 p.p.m. In the declining South Carolina colonies eight of the eleven eggs collected in 1969 and two of ten collected in 1970 contained DDE concentrations greater than 4 p.p.m. Thirteen of the twenty-one eggs collected in the 2 yr exceeded 15% shell thinning. In California, where reproductive failure occurred, a 35% decrease in eggshell thickness was associated with 71 p.p.m. of DDE in the egg.

In contrast, only ten of the forty-nine brown pelican eggs collected in Florida were more than 15% thinner than the pre-1947 average; and residues exceeded 4 p.p.m. in only four eggs. It seems probable, therefore, that some pelicans in Florida are adversely affected by shell thinning although the extent of this thinning is not great enough for obvious effects on the size of the populations.

In contrast to the brown pelican, the herring gull (*Larus argentatus*) showed no thinning when DDE residues were between 4 and 5 p.p.m. and only 11% thinning when residues were near 80 p.p.m.⁸. The eggshell thinning response induced by DDE in the brown pelican was similar to that found in the prairie falcon⁵. The double-crested cormorant⁴ seemed somewhat more resistant to thinning by DDE than these two species, but shell thinning generally occurred in cormorant eggs that contained less than 5 p.p.m. of DDE and 15% thinning was

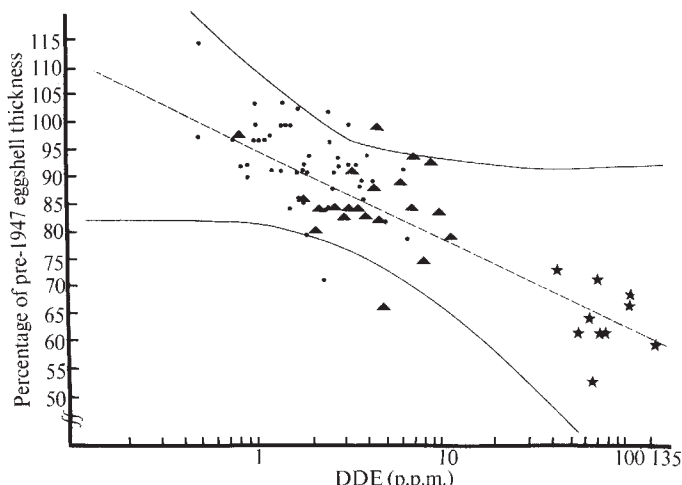


Fig. 1 Association of DDE residues in eighty brown pelican eggs from Florida (●), South Carolina (▲) and California (★) with the % of pre-1947 eggshell thickness. Solid lines represent 95% confidence limits. $\bar{Y} = 95.787 - 15.689 \log_{10} X$; $r = -0.80$ ($P < 0.01$).

recorded in one colony when DDE egg residues averaged just under 20 p.p.m.

The concentration-effect relationship involving the logarithm of DDE and eggshell thinning seems to exist and to follow a mathematically similar pattern in different species, but to operate at different levels in different species. A level of DDE in eggs that would result in population collapse among brown pelicans would not be expected to effect an overall population change among herring gulls.

We thank the many individuals who helped us with the collection of eggs.

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LETTERS TO NATURE

PHYSICAL SCIENCES

Detection of Radio Emission from GX9+1

VARIABLE radio sources have been reported¹⁻⁵ to be associated with the X-ray sources Sco X-1, Cyg X-1 and GX17+2. We report here the detection of a variable radio source in association with the X-ray source GX9+1.

The observations of GX9+1 were made with the NRAO three-element interferometer at frequencies of 2,695 and 8,085 MHz. Previous negative results for radio emission from this X-ray source, together with a description of the interferometer and data analysis methods, are given elsewhere³. Observations taken on May 29, May 31 and November 8, 1971, placed an upper limit of $5 \times 10^{-29} \text{ W m}^{-2} \text{ Hz}^{-1}$ on the flux density for any steady point radio source within 10 arc min of the X-ray position (ref. 6):

$$\alpha_{1950} = 17 \text{ h } 58 \text{ m } 35 \text{ s} \pm 6.8 \text{ s}$$

$$\delta_{1950} = -20^\circ 31' 30'' \pm 1.7''$$

On November 6, 1971, a radio source with a mean flux density of $(8 \pm 3) \times 10^{-29} \text{ W m}^{-2} \text{ Hz}^{-1}$ at 2,695 MHz appeared at the following position:

$$\alpha_{1950} = 17 \text{ h } 58 \text{ m } 34.0 \text{ s} \pm 0.3 \text{ s}$$

$$\delta_{1950} = -20^\circ 30' 58'' \pm 5''$$

This radio source appears unresolved at all spacings and must therefore be smaller than one arc s. Note that the detection and position determination of the radio source are based on the phase data rather than the amplitude data (flux density). The latter are much more uncertain at weak flux levels³. No radio source was detectable at 8,085 MHz during this time, with an upper limit of $5 \times 10^{-29} \text{ W m}^{-2} \text{ Hz}^{-1}$. At the time the radio source flared above the detection limit it was being tracked during alternate half hours over a period of about 7 h. Although the source was clearly present during

all of this 7 h period, variations of a factor of two about the mean strength could not be distinguished.

The variability of this radio source is similar to that observed for Sco X-1 (refs. 2, 5) and GX17+2 (ref. 3). This fact, together with the excellent agreement with the X-ray position, indicates the probable detection of radio emission from GX9+1. The nonthermal spectral index for the source during the flaring period further suggests that the object is more closely related to Sco X-1 (refs. 2, 5) than Cyg X-1 (refs. 3, 7).

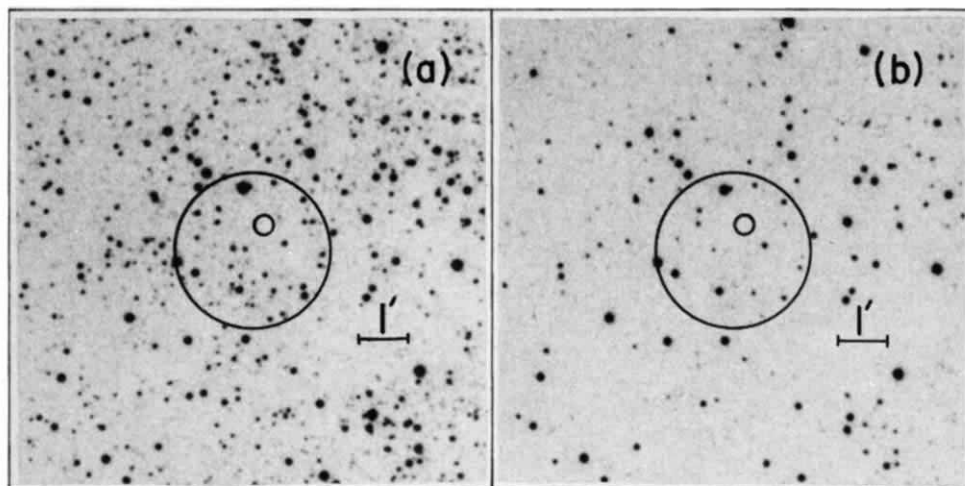
Previously, a search for the optical counterpart of GX9+1 (ref. 8) down to $B=17$ within the X-ray error circle (9 square arc min)⁶ had not yielded any likely candidates. With the present smaller region of uncertainty (0.02 square arc min) the search for an optical counterpart can be extended to fainter stars.

Fig. 1 shows enlarged portions of the red and blue *Palomar Sky Survey* prints of the region near GX9+1. There is no stellar-like object at the location of the radio source (small circle, Fig. 1) down to the print limits. If we use Sco X-1 as a model and assume that the optical flux scales proportionally to the X-ray⁹ and the radio-flare^{2,5} fluxes, the unreddened visual magnitude of the optical counterpart of GX9+1 should be about 16.5. The lack of any visible object implies that either there is considerable visual obscuration in the direction of GX9+1 or the optical radiation is relatively weaker than for Sco X-1.

Two other celestial X-ray sources, GX349+2 and GX340+0, whose positions have been well determined¹⁰, were also observed for radio emission during the same period of observations of GX9+1. These two sources should be good candidates for radio emission because they are members of the complex of relatively stable X-ray sources, along the galactic plane and in the vicinity of the galactic centre, which includes GX9+1 and GX17+2. The present observations set an upper limit of $5 \times 10^{-29} \text{ W m}^{-2} \text{ Hz}^{-1}$ at 2,695 and 8,085 MHz for any steady point radio sources near these two X-ray sources. Continued searches should be made, however, in view of the likelihood of highly variable radio emission.

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Fig. 1 Enlargements of the red (a) and blue (b) *Palomar Sky Survey* prints (copyright National Geographic Society) of the region near GX9+1. North is up and east is to the left. The large circle in each print represents the X-ray source location (3σ) (ref. 6). The small circle encloses the radio source location and, for clarity, is drawn twice the size of the quoted uncertainty.



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IMPORTANT information in astronomy may be gained in the not-too-distant future by the observation of gravitational waves. But the nonlinearity of the gravitational field equations in general relativity makes it difficult to estimate effects which might arise through collisions between waves, even when the nature of the impinging waves is known. In general, direct superposition of solutions for individual waves will lead to violation of the vacuum equations. One thus has to determine, in a particular situation, whether there exists a valid vacuum solution, non-singular in the wave region, which represents the gravitational field before, during and after the collision, or whether instead the eventual formation of physical singularities is predicted. For example, Penrose¹ has shown that in the collision of (exact) weak "sandwich" plane waves², each will warp the other until singularities occur in the wave-front.

In a further investigation³, Penrose and Khan studied an exact solution for colliding sandwich waves using a form of metric due to Rosen. Because these waves are "sandwiched" between flat regions of space-time, the superposition of solutions prior to a head-on collision is straightforward. For parallel-polarized impulsive plane waves, with delta-function amplitudes, they were able to show that the planeness of the waves is not retained, and that the solution can be followed through the collision until the inevitable occurrence of a physical singularity, due to focusing. This raises the question of whether too sharp an amplitude-variation gives rise to singularities in a less highly symmetric model. And if so, how sharp the variation may be if singularities are not to occur.

Fig. 1 Heavy arcs show the wave region of a smooth gravitational pulse in a meridian section $t=\text{constant}$ ($t>b>0$), $\varphi=\text{constant}$, the pulse having emanated from the ring $Q(r=b, z=0)$ at $t=0$. Self-reflexion has occurred on $r=0$ at $t=b$, as indicated by the arc EF .

solution, although somewhat specialized, can give insight into collision processes in other realistic models because the radiation zone here is bounded. If time-symmetry is assumed, the solution is non-singular everywhere (even on the ring), and is then a toroidal analogue of the smooth imploding-exploding cylindrical wave of Weber and Wheeler⁴. I consider here only the outgoing ($t > 0$) phase, where $t = 0$ is the moment of time-symmetry.

The solution is constructed by bending an imploding-exploding cylindrical wave (such as Weber and Wheeler's) by introducing "bending terms"^{5,6} into Rosen's metric⁷

$$ds^2 = e^{2(\gamma-\psi)} (dT^2 - dR^2) - R^2 e^{-2\psi} d\phi^2 - e^{2\psi} dZ^2 \quad (1)$$

where ψ , γ are functions of R , T only, and ψ satisfies, *in vacuo*, a linear wave equation. Once ψ is chosen the remaining vacuum equations determine γ . The bending terms have the property that if the wave amplitude is set equal to zero the space-time is flat, whether or not the terms are included, whereas for arbitrarily small non-zero amplitudes the wavefronts are totally different in form in the two cases. Bending terms for (1) are

$$\begin{aligned}\psi &= \psi_0 = \frac{1}{2} \ln [T + (T^2 - R^2)^{\frac{1}{2}}], \\ \gamma &= \gamma_0 = \psi_0 - \frac{1}{2} \ln [2(T^2 - R^2)^{\frac{1}{2}}]\end{aligned}\quad (2)$$

the flat solution (1), (2) being obtained from the Minkowski metric in cylindrical polar coordinates (r, φ, z) and time t by the transformation

$$T = \frac{1}{2}(r^2 + t^2 - z^2), \quad R = r(t^2 - z^2)^{\frac{1}{2}}, \quad Z = \tanh^{-1}(z/t) \quad (3)$$

If $\psi = \alpha(R, T)$ represents any cylindrical wave solution of (1), then $\psi = \psi_0 + \alpha$ defines a new solution, although only in a few cases is the latter of physical interest or free of unphysical singularities. The Weber-Wheeler wave is given by

$$\psi = \alpha = A \operatorname{Re}\{[a - i(T - \frac{1}{2}b^2)]^2 + R^2\}^{-1/2} \quad (4)$$

where a, b are real constants, and is time-symmetric about $T = \frac{1}{2} b^2$. The main part of the pulse is confined near $T \pm R$

$=\frac{1}{2}b^2 (R \geq 0)$, and both incoming and outgoing phases are used to construct the outgoing toroidal wave.

Consider (3) as a one-one mapping of the region $M_1[t > (r^2 + z^2)^{1/2}, r > 0]$ onto the region $T > R > 0$. The locus $T - R = \frac{1}{2}b^2$ corresponds, in each meridian 2-space $t = \text{constant}$, $\varphi = \text{constant}$ to an arc of the circle $(r+b)^2 + z^2 = t^2$. In the interesting case $t > b > 0$, this has real points in M_1 as shown in Fig. 1 by the arc EF , while $T + R = \frac{1}{2}b^2$ corresponds to arcs of the circle $(r-b)^2 + z^2 = t^2$ (CE and DF). In the same way (3) can be used as a one-one mapping of the region $M_2[|z| < t < (r^2 + z^2)^{1/2}, r > 0]$ onto $T > R > 0$, so that $T - R = \frac{1}{2}b^2$ corresponds to the semicircular arc AB and $T + R = \frac{1}{2}b^2$ to arcs AC and BD .

Taking $\psi = \psi_0 + \alpha$, with α given by (4), we get, on applying the inverse transformation to (3), a solution in $|z| < t$ for a pulse wave mainly confined near the locus represented by solid arcs in Fig. 1. When A is small the space-time region is nearly flat and the coordinates (r, φ, z) continue to resemble cylindrical polars. The indentation EF represents the wave due to self-reflexion on $r=0$, and is not present for $0 < t < b$, when the corresponding diagram for the pulse is a circle of radius t and centre $Q(r=b, z=0)$. An extension to $|z| > t$ is possible with use of an analogous transformation to (3).

The main result to emerge is that the character of the wave for this choice of α remains the same indefinitely, so that there is no significant focusing effect. Another feature of this (time-symmetric) solution is that the distant field "due" to the bounded region of radiation can be studied; it is found to resemble that of a source with a form of octupole structure. It would, of course, be interesting to compare these results with those for less smoothly varying functions α .

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Uptake of Cobalt from Seawater by Aeolian Dust

UPTAKE onto the surfaces of detrital particles has been considered as a possible mechanism for the removal of trace elements from seawater^{1,2}. But Kharkar *et al.*³ have examined experimentally the adsorption of trace elements by detritus in both river and marine environments. They concluded that when a trace element is adsorbed from solution in river water it is always released, to a greater or lesser extent, on contact with seawater. While they agreed with earlier findings^{1,2} that detritus placed in seawater can remove trace elements from it, they postulated that this does not happen in the transport of river detritus to the oceans.

It seems therefore that a critical factor controlling the uptake of trace elements from seawater by detritus is the sedimentary pathway by which the detritus reaches the oceans. Further, detritus which has been brought to the oceans in a "dry" state may behave differently with respect to trace element uptake than that brought by rivers.

There have been various detailed studies⁴⁻⁸ of the distribution of aeolian material in the atmosphere over deep-sea areas, and because this material is introduced directly to seawater in a "dry" state it appeared to us that it might remove trace elements from seawater. Here we report the initial results of experiments designed to examine the possibility of such uptake.

Cobalt was chosen as a test element because it has been shown to be partially desorbed from river detritus on contact with a saline environment³. Its uptake from seawater by north-east trade wind dust was examined.

To investigate the uptake of Co at the low concentrations present in seawater ($< 0.2 \mu\text{g/l}$) it was necessary to use a radio-tracer technique. Uptake measured in this manner will, however, include any exchange which occurs between ^{60}Co in solution and stable Co on the surface of the dust. This kind of exchange will not result in the net loss of total Co from the solution, and true uptake can only be measured by determining the concentration of total Co in solution before and after reaction with the dust. Such determinations are extremely difficult at the low concentrations of Co found in seawater. To establish therefore that direct (or non-exchange) uptake of Co occurs when the dusts are in contact with seawater the reactions between dust samples and seawater containing added Co were investigated using a polarographic technique⁹ (analyst, L. G. Royle). The results indicated that in the conditions used the dusts removed an average of 70% of the Co (range 60–80%) from the seawater. A parallel experiment was run in the same conditions but with the addition of ^{60}Co to the seawater, and the Co uptake measured as a function of the activities of ^{60}Co was roughly 60%. It is evident therefore that the dusts can take up Co directly from seawater. Because of the reasonable similarity between the results of the experiments using stable and radioactive Co it is assumed that the use of ^{60}Co will give, to a first approximation, an estimate of the amount of uptake of stable Co from seawater in the conditions used. In all subsequent experiments therefore ^{60}Co was used in the conditions described below.

Sufficient ^{60}Co radio-tracer (as CoCl_2) to give an adequate counting level was added to a stock sample of seawater (salinity roughly 32‰ filtered through a $0.45 \mu\text{m}$ filter). The ^{60}Co solution contained a small amount of trace element carrier but the total addition of Co from this source was less than that normally present in seawater. In each experimental run 50 ml. of the stock seawater was pipetted into four 125 ml. capacity glass bottles. To the stock seawater in three of the bottles an amount of aeolian dust was added, in the range 0.1 to 0.005 g (Table 1), and the bottles were shaken mechanically (to prevent particles from settling) at a particular temperature in a thermostatically controlled water bath; the ranges of temperatures used were 8° C, 10° C, 15° C and 20° C. The fourth bottle, which did not contain a dust sample, was also shaken to act as a control on the uptake of ^{60}Co onto the glass walls of the bottles; this uptake was found to be negligible in all experimental runs. It was initially established that the uptake of ^{60}Co onto the dusts had reached a maximum after roughly 48 h and all the reported measurements were taken after the suspensions had been shaken for longer than this time. At this stage aliquots were withdrawn from the seawater-dust suspensions and centrifuged. A portion of the clear supernatant liquid was withdrawn and its activity measured on an EKCO N664A scintillation counter and an EKCO N530F scaler. The activity of each aliquot was compared with a similar aliquot of liquid from the control bottle, and the difference between the two activities was assumed to be a measure of the uptake of ^{60}Co by the dusts.

Table 1 shows that these types of aeolian dusts can remove ^{60}Co from seawater within the temperature and pH ranges expected in the average 4,000 m deep-seawater column. These initial experiments were designed simply to establish whether or not natural aeolian dust can remove trace elements from seawater. We felt this to be necessary because aeolian dust represents a genetically different type of detritus from that brought to the sea by rivers, that is, the former may be regarded as being in a "dry" state and the latter in a "wet" state. This means that it is incorrect to assume that laboratory experiments^{1,2} in which previously dried detritus is allowed to react with trace elements dissolved in seawater are directly applicable to all actual uptake processes which occur in the oceans,

Table 1 Experimental Uptake of ^{60}Co from Seawater by Aeolian Dusts

Dust sample*	Sample locations†	Content of organic C (wt %) [‡]	Amount of dust/50 ml. seawater (g)	pH of seawater Initial	pH of seawater Final	Temperature (°C)	% removal of ^{60}Co from seawater§	Remarks
GH5	12° 27' N, 17° 28' W 20° 20' N, 17° 43' W	0.94	0.1	7.7	7.5	20	36	
			0.1	7.9	7.8	20	46	
			0.05	7.9	7.9	20	33	
			0.01	7.9	8.0	20	4.5	
			0.005	7.9	7.9	20	5.0	
			0.1	One series, initial pH 7.7 final pH 7.5		15	40	
			0.1			10	40	
			0.1			8	44	
HG27	18° 30' N, 24° 10' W 20° 19' N, 22° 33' W	1.4	0.1	One series, initial pH 7.7 final pH 7.5		20	37	
			0.1			15	38	
			0.1			10	38	
			0.1			8	31	
M2	18° 55' N, 17° 22' W 20° 46' N, 18° 49' W	2.7	0.1	7.6	—	20	30	
M3	20° 46' N, 18° 49' W 20° 47' N, 17° 46' W	3.5	0.1	7.8	—	20	35	
HG25	13° 09' N, 26° 44' W 15° 34' N, 26° 03' W	—	0.1	8.0	—	20	56	
M19	33° 19' N, 09° 42' W	—	0.1	8.0	—	20	15	This sample was heavily contaminated with ship smoke
		—	0.1	8.0	—	20		

* The sample numbers refer to the cruise on which the collections were made.

† The samples were collected on board ship by a mesh technique; the coordinates refer to the start and finish of each collection.

‡ Organic carbon was determined by Dr S. E. Calvert.

§ The uptake is reported as a percentage of the total ^{60}Co originally in solution; each result is the average of at least two experimental runs.

because the detritus has not passed through the experimental equivalent of a river environment. Such experiments may, however, be applicable to material brought to the oceans in a "dry" state, for example, from aeolian transport.

It is probable that the removal of Co from seawater onto the dusts occurs by some form of surface adsorption, for example, specific exchange involving the species Co^{2+} and ions held on the surface of the clay minerals or iron oxides^{6,7} constituting the dusts. Organic material can act as an adsorbant for trace elements from seawater. The dusts used in the present experiments, however, contained only small amounts of organic carbon (Table 1), and it is unlikely that the organic material was responsible for the removal of Co from seawater.

There are many inherent difficulties in attempting to reproduce open-ocean conditions in the laboratory, and so we have not attempted to extrapolate our experimental results to processes controlling the trace element geochemistry of deep-sea sediments. Much more sophisticated experiments must be designed before this can be done. We simply suggest that aeolian material, which is introduced into seawater in a "dry" state, is capable of at least the short term uptake of Co (and by analogy other trace elements) from the upper few metres of ocean water.

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Electrostatic Hazards in Supertanker Cleaning Operations

ELECTRIC fields of up to 300 kV m⁻¹ have been measured in the centre tanks of 200,000 ton tankers (very large crude carriers) (VLCC) during normal cleaning. The tanks are cleaned by means of high velocity jets of water having flow rates of up to 180 m³/h and velocities of up to 40 m/s at the nozzle. Disintegration of the water jet at the tank walls gives rise to a cloud of charged water droplets. The coarser drops resulting from splashing all assume the same sign of charge, while the particles in the finer mist retain the opposite sign¹. The larger drops precipitate more rapidly than the finer mist, and this gravitational sorting results in an electric field. Near natural waterfalls fields of the order of 100 V/m have been reported^{1,2}.

It is not yet known, however, whether this electric field is a potential hazard in the cleaning of tankers. With the abundance of earthed protrusions (for example, walkways, stairways, strengthening structures) within the cargo tanks and the gun cleaning nozzle penetrating 5 m below deck level, conditions favourable for discharges may well be created as a result of local field intensification. Such conditions are a possible ignition hazard.

I have concentrated on the importance of the presence of an earthed protrusion in a charged water aerosol, using an experimental arrangement comprising a polyethylene enclosure, dimension approximately 2 m × 2 m × 3 m, into which a highly charged water aerosol was injected. A modified form of electrostatic airless spray gun was used with a variable HT generator of 0–100 kV. The aerosol particle size was of the order 10–20 μm diameter, and charging was effected by means of corona points mounted close to the nozzle.

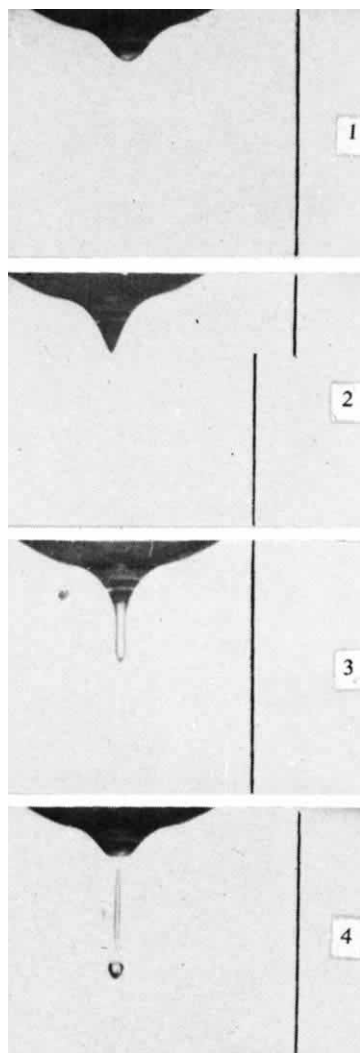


Fig. 1 Falling water filament and signal*. (500 p.p.s. signal line drawn in for clarity.) 1, Filament beginning to fall; 2, appearance of pointed head, onset of signal; 3, filament extending; 4, filament disconnected, disappearance of signal.

* Non-consecutive frames. Number of frames between 1 and 4=34.

A metal spherical electrode, diameter 3.7 cm, mounted on a boom was used as an earthed test probe which could be placed at any position in the aerosol cloud. With this simple arrangement, the earthed probe was to represent any earthed protrusion within a VLCC—such as the gun cleaning nozzle itself. I attempted to simulate, very crudely, the conditions which might apply in the case of an earthed object in the large tanks of a VLCC by establishing a value of electric field in the small laboratory tank similar in value to that measured in the VLCC. This was achieved by establishing a much higher charge density in the laboratory tank than is measured in the VLCC tank ($\approx 10^{-6}$ C/m³ as compared with 10^{-8} C/m³ in the ship).

Charging currents from the spray gun equipment were typically of the order of a few hundred microamperes with the corona points held negative, while the electric fields within the enclosure were in the region of 40–50 kV/m. Fields were measured using the Langmuir probe technique, hence the lower values than previously quoted for fields within the cargo tanks which were measured with a conventional field mill. Intensification of the field caused by the presence of the field mill introduces this inevitable error. On introducing the earthed spherical electrode into the charged mist, the sphere very quickly accumulated a coating of water—wetting in this case being aided very effectively by the charged water droplets'

natural tendency to migrate towards an earthed object. The electrode was connected directly into an oscilloscope, and signal pulses indicative of small bursts of microdischarges were recorded. The visible discharge and electrode signal occurred simultaneously. Closer observation of the sphere and analysis of water drops falling from it revealed a narrow, elongated drop profile. High speed movie photography at various speeds up to 3,000 frames per second permitted detailed observation of the drop history. Fig. 1 shows the sequence of events for a typical drop falling in a high field. Simultaneously recorded on the film is the oscilloscope signal associated with departure of drops off the electrode, which clearly shows the signal appearing only when the leading point of the water filament assumes a certain critical radius of curvature. The signal disappears at the instant the elongated water filament breaks contact with the sphere. Observation and the exact positioning of the visible discharge were accomplished using a standard image intensifier tube. Fig. 2 illustrates a typical discharge appearing at the pointed end of a falling water filament.

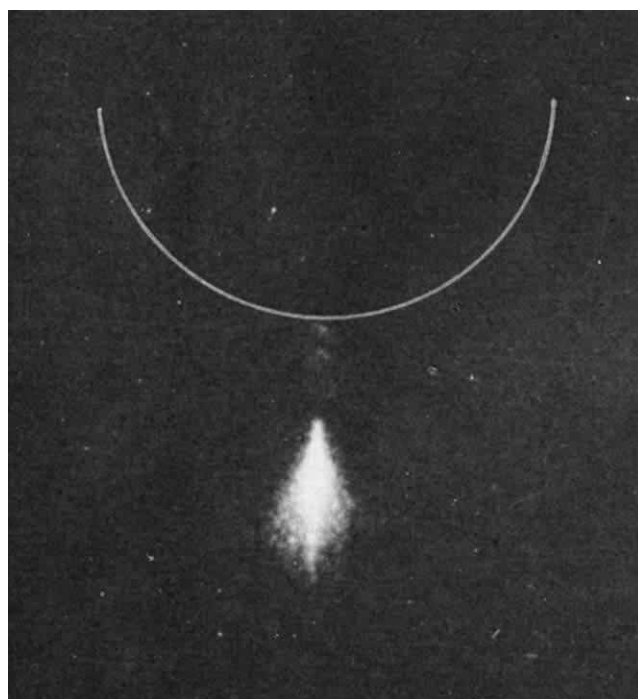


Fig. 2 Image intensifier photograph of discharge from falling water filament.

I suggest that these visible discharges are in fact corona pulses from the pointed end of the falling water filament, typical of the events noticed and studied in detail by English³.

Because dripping water within the structure of a VLCC is an obvious occurrence, and such high electric fields have been measured within the tanks during washing, further investigation of such discharges warrants serious consideration. It is interesting to note that the phenomenon is in fact very similar to the electrostatic detearing process used in painting. Preliminary tests have been conducted in attempts to determine the energy involved in the discharges, and results to date show that under certain conditions these discharges are indeed capable of igniting various explosive mixtures. Much further work is necessary, however, before a precise energy value can be safely quoted. The nature of the water used is another avenue for further investigation because freshwater, seawater with or without detergent, or possibly aerosol particles comprising a cleaning fluid enclosed in an "oily" envelope, could all behave differently in their eventual ignitability characteristics.

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Are Microtektites the Result of Cometary Impacts with the Earth?

Durrani and Khan¹ suggest that microtektites obtained from ocean sediments near Australia and the Ivory Coast may be the result of impacts of comets with the Earth. They measured the ages of the microtektites obtained from the two regions and determined the ages at 0.71 ± 0.1 and 0.88 ± 0.13 m.y., respectively, for Australasian and Ivory Coast tektites. These ages agree closely with the dates of two of the reversals of the geomagnetic field, and this close agreement is interpreted by them as suggesting that the microtektites and magnetic reversals have resulted from impacts of comets with the Earth.

Interesting as the idea may be, we wish to point out that there seems to be a difficulty regarding the frequency of cometary impacts with the Earth. The frequency was estimated by Russel, Dugan and Stewart and it seems best to quote their statement. They write²: "It may readily be computed that a small rapidly moving body which approaches the Sun within one astronomical unit stands about one chance in 400 million of hitting the Earth. Because roughly five comets come within this distance every year, the nucleus of a comet should hit the Earth, on the average, once in about 80 million years. Collisions with the outer parts of the head should be many times more frequent".

For the suggestion by Durrani and Khan to be valid, the frequency of the cometary impact would have to be about one in a million years, while the frequency given by Russel *et al.* is some 80 times smaller. It is true that not all comets that come within one astronomical unit from the Sun are detected. Nevertheless, it seems that the discrepancy by the factor of 80 cannot easily be explained by taking into account comets that escape detection.

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¹ Durrani, S. A., and Khan, H. A., *Nature*, **232**, 320 (1971).

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DR DURRANI writes: If the orbits of approaching comets were completely random, the probability¹ that one of them, having arrived at about 1 AU (Earth-Sun distance) from the Sun, would hit the Earth would be related to the solid angle presented by the Earth, that is, $\pi r^2/4\pi R^2$, where r is the radius of the Earth and R is the Earth-Sun distance. Certain additional factors must, however, be taken into account: first, a factor of 2 to allow for collisions during the return journey of the comet; second, the Earth's influence in deflecting a passing comet (which depends on the initial velocity and orbit of the comet); this increases the collision cross section by a factor of about 3 (ref. 2). On taking these factors into account, the probability of a comet at 1 AU from the Sun hitting the

Earth is seen to be approximately 1 in 360 million (which is not far off the estimate given in ref. 3 and quoted by Drs Kellner and Yabushita).

The number of comets arriving in the vicinity of the Sun, however, is taken by Urey¹ to be 10 per year, as opposed to 5 per year assumed by Russel *et al.*³ Urey's estimate leads to a collision probability per year of 1 in 36 million. As Kellner and Yabushita point out, not all comets that come to within 1 AU of the Sun are detected. Indeed, according to Russel *et al.* (ref. 3, page 425), "If allowance is made for the fact that comets of smaller perihelion distances may be unfavourably placed and so miss discovery, it is probable that not more than 1/8 of those which come within 5 astronomical units of the Sun are picked up". Thus, one would not be surprised if the above figure of 1 in 36 million were increased by a factor of 3 or 4 (to 1 collision in, say, every 10 m.y.). Urey⁴ quotes a personal communication from Z. Kopal to the effect that one collision of a comet head with the Earth every 50 million years "was much too low an estimate of such events".

With the above conclusions in mind, I shall examine the data. Four distinct tektite falls have been observed during the past 35 million years (Australasian, approximately 0.7 m.y.; Ivory Coast, 0.9 m.y.; Czechoslovakian, 14 m.y.; and North American, 35 m.y.). Cometary collisions are such rare events that they obey Poisson distribution. What is the probability, then, of observing four falls during the past 35 million years if the expected frequency was (a) 1; (b) 2; (c) 3 and (d) 4, as indicated above? The answers are: (a) 1.5%; (b) 9.0%; (c) 16.9%; and (d) 19.5%. (Note that even when four falls were expected over the 35 m.y., the probability of actually finding nothing but four falls is given, by Poisson distribution, as roughly 20%.) Thus the probability of finding four tektite falls through cometary collisions over the past 35 m.y. is (contrary to the expectation of Drs Kellner and Yabushita) not a negligible quantity even when the expected frequency was only one or two over this period.

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BIOLOGICAL SCIENCES

Adenylic Acid-rich Sequence in RNAs of Rous Sarcoma Virus and Rauscher Mouse Leukaemia Virus

ADENYLIC ACID (A)-rich sequences have been found in some mRNAs of animal cells^{1,2} and DNA viruses³ but not in the RNAs of cytocidal or oncogenic RNA viruses. These sequences have been proposed to be involved in the transport of mRNA to the cytoplasm from the nucleus or vaccinia virus core^{2,3}, or in the association of mRNA with proteins necessary for translation¹.

In certain conditions, however, the DNA polymerase of Rous sarcoma virus (RSV) transcribes a T-rich polynucleotide from RSV RNA⁴, implying that an A-rich region exists in RSV RNA⁴. Previous hybridization experiments by Harel *et al.*⁵ with 60-70S RNA of avian myeloblastosis virus and chicken

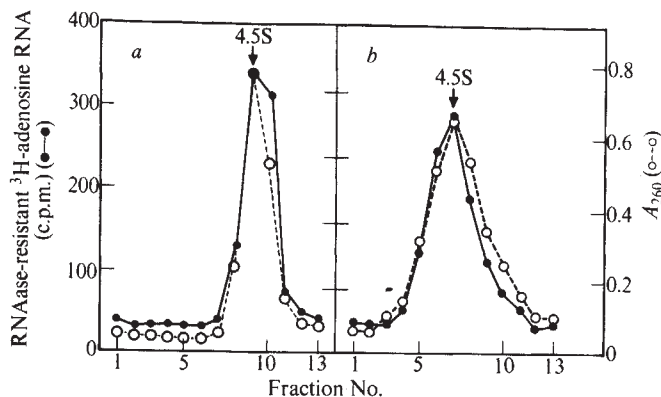


Fig. 1 Zonal sedimentation of the RNAase-resistant fraction of ³H-A-labelled RSV RNA (a) or MLV RNA (b). ³H-A-labelled 60-70S RSV RNA or MLV RNA was digested exhaustively with pancreatic and T₁ RNAase in 0.3 M NaCl (see Table 1). The digest was phenol extracted three times in the presence of 1% sodium dodecylsulphate (SDS) and then precipitated with two volumes of ethanol. After washing with 75% ethanol twice the precipitate was dissolved in standard buffer (0.01 M Tris, pH 7.4, 0.1 M NaCl and 1 mM EDTA) containing 0.2% SDS. Sedimentation was in a 10%-25% sucrose gradient containing 0.1% SDS and standard buffer in a Spinco SW65 rotor at 65,000 r.p.m. at 20° C for 6 h (a) or for 8 h (b); 100 μg tRNA was included as a marker. Fractionation and analysis of the gradients were as described previously^{8,9}.

DNA have also suggested that there may be an A-rich sequence in the viral RNA. The evidence, however, that an A-rich sequence of avian myeloblastosis virus RNA is virus-specific was not compelling since only 2-4% of the viral RNA hybridized with cellular DNA, and the virus has been shown to contain cellular RNAs⁷. We report here direct evidence that there are A-rich sequences in 60-70S RSV RNA and Rauscher mouse leukaemia virus (MLV) RNA.

The Prague strain of RSV of subgroup C was used in all experiments. Growth of virus and purification of the viral RNA followed published procedures⁹. Rauscher MLV was propagated in the JLS V5 cell line²⁶ and influenza virus (WSN) in a bovine kidney cell line²⁷. Labelling of the viral RNA with adenosine-2-³H (10.6 Ci/mmol., New England Nuclear) was in 10 cm Petri dishes with 6 ml. Dulbecco's modified Eagle's medium (Grand Island Biologicals) supplemented as described previously⁹ and containing 50-100 μCi, for periods between 3 and 12 h.

To determine whether an A-rich sequence is associated with 60-70S RSV RNA, we have exhaustively digested the RNA with pancreatic and T₁ RNAase. Poly A has been shown by Beers⁶ to be RNAase-resistant at high ionic strength, although it becomes RNAase-sensitive at low ionic strength. We have confirmed these findings using ³H-poly A (Miles Laboratory). As shown in Table 1, about 9% of the adenylic acid in ³H-A

labelled 60-70S RSV RNA is RNAase-resistant in 0.3 M NaCl. By contrast less than 1% of the radioactivity of 60-70S ³H-uridine-RSV RNA is RNAase-resistant in the same conditions. This RNAase-resistant fraction of ³H-A 60-70S RSV RNA is insensitive to pronase and DNAase but is hydrolysed by KOH or by RNAase at low ionic strength (Table 1). We conclude from these results that 9% of the adenosine of RSV RNA may be in an RNAase-resistant, A-rich sequence.

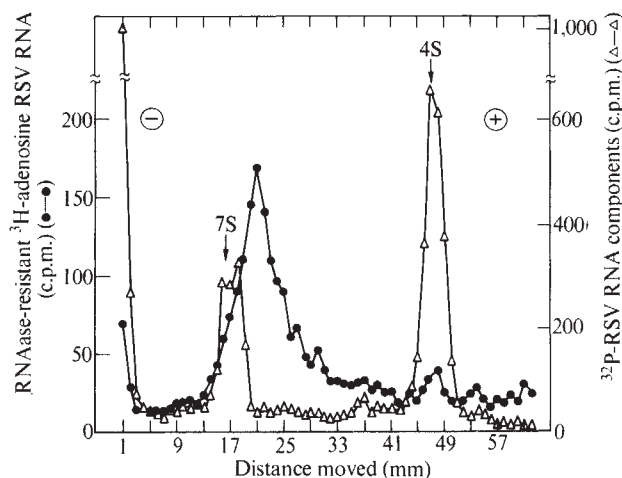


Fig. 2 Polyacrylamide gel electrophoresis of the RNAase-resistant fraction of ³H-A-labelled 60-70S RSV RNA. The RNAase-resistant fraction of ³H-A-labelled 60-70S RSV RNA was prepared as described for Fig. 1. Before electrophoresis it was dissolved together with low molecular weight fractions of ³²P-RSV RNA in electrophoresis sample buffer²⁴. Electrophoresis was in a 10% diacrylate-crosslinked polyacrylamide gel (6×0.7 cm) at 50 V for 220 min²⁴. Methylene blue was included as a marker. It moved at the same rate as 4S RSV RNA.

Assuming that the A-content of RSV RNA is 25%²², this conclusion agrees with the finding that 2.5% of the radioactivity of 60-70S ³²P-RSV RNA consists of an RNAase-resistant (Table 1) fraction containing 95-97% adenylic acid (Table 2). This A-rich sequence has about the same sedimentation coefficient as tRNA (Fig. 1), which is 4.5S¹⁰. Using the formula $s_{20,w} = 0.405Z^{0.467}$ (Z is the degree of polymerization)¹¹ and assuming that it is poly A, we have estimated its molecular weight to be about 60,000. During polyacrylamide gel electrophoresis, however, it moves farther than the 7S component of RSV RNA^{7,12} but considerably less than the tRNA component of RSV¹⁵ (Fig. 2). The low electrophoretic mobility and the low sedimentation coefficient of the A-rich fraction of RSV RNA relative to those of the

Table 1 Effects of Various Treatments on Fractions of RSV RNA

Treatments	60-70S RSV RNA			4-12S RSV RNA	
	³ H-A	³² P	³ H-U	³ H-A	³ H-U
None	100	100	100	100	100
Pancreatic RNAase+RNAase T ₁	9±1	2.5±0.5	0.7±0.2	7±1	0.6±0.2
Pancreatic RNAase+RNAase T ₁ +DNAase	8±1	—	—	6±1	—
KOH	1±0.5	—	0.5±0.2	1±0.5	—
Pronase	100±1	—	—	100±2	—
Pancreatic RNAase+RNAase T ₁ at low ionic strength	1±0.2	—	—	—	—

The numbers represent the average percentage of 5% trichloroacetic acid-precipitable fractions of radioactive RNA obtained after two to four determinations (± range) following the treatments indicated. RNA samples (2,000-4,000 c.p.m.) in 0.01 M Tris, pH 7.4, 2 mM EDTA were heated at 100° C for 45 s and quenched at 0° C before nuclease digestion. The standard digestion of RNAs, which was used in all experiments unless stated otherwise, was with pancreatic RNAase (20 μg/ml.) and RNAase T₁ (150 U/ml.) in 0.3 M NaCl, 0.03 M Na citrate, pH 7.0, for 30 min at 38° C. RNAase (as above) and DNAase (20 μg/ml.) digestion was in 0.01 M Tris, pH 7.4, 6 mM MgCl₂, 0.05 M NaCl for 30 min at 37° C²³. Treatment with 0.2 M KOH was for 18 h at room temperature. Pronase (1 mg/ml.) digestion was for 48 h in 0.01 M Tris, pH 7.8, at 38° C. Digestion with pancreatic RNAase (20 μg/ml.) and RNAase T₁ (150 U/ml.) at low ionic strength was in 0.01 M Tris, pH 7.4, and 2 mM EDTA for 30 min at 38° C.

tRNA marker, of molecular weight 25,000, are presumably due to its single-stranded helical structure which is typical of poly A^{11,13,14}.

The slowly sedimenting (4-12S) fraction of ³H-A-RSV RNA obtained after sucrose gradient sedimentation of total viral RNA also contains an A-rich sequence (Tables 1 and 3). This is indistinguishable from that of the 60-70S RNA by sucrose gradient sedimentation or by polyacrylamide gel electrophoresis (not shown). This result is compatible with the view that some of the 4-12S RSV RNA derives from degradation of 60-70S RNA^{16,20,22}.

Table 2 Base Composition of the RNAase-resistant Fraction of 60-70S ³²P-RSV RNA

	Mol percentage of ³² P-nucleotides			
	C	A	G	U
Preparation A	<0.5	97	2	<0.5
Preparation B	2	95	3	<0.5

60-70S ³²P-RNA was prepared as described previously⁴ and digested with RNAase in 0.3 M NaCl (Table 1). After three phenol extractions (Fig. 1), the digest was twice precipitated with two volumes of ethanol in presence of carrier TMV RNA. The digest was freed of oligonucleotides either by binding to 'Millipore' and subsequent washing (Table 3) (A) or by precipitation with 2.5% perchloric acid (B). The RNAase-resistant ³²P-RNA (3,000-5,000 c.p.m.) was then mixed with 200 µg TMV RNA and incubated in 0.2 ml. 0.3 N KOH for 18 h at 37° C (ref. 25). (If ³²P-RNA bound to 'Millipore' filter was hydrolysed, an additional incubation with an extra 0.2 ml. 0.3 N KOH for 12 h was necessary to obtain complete hydrolysis. Presumably hydrolysis of RNA was delayed because KOH reacted with the 'Millipore' filter.) Electrophoresis was on 'Whatman 3MM' paper in 0.04 M Na-citrate, pH 3.5, for 2 h at 15° C and 40 V/cm²⁵. Unlabelled nucleotides were located visually by ultraviolet absorbance. The ³²P-nucleotides were detected by determining the radioactivity of 0.5 cm strips of the pherogram in toluene based scintillation fluid.

The 60-70S RNA of RSV has a complex subunit structure^{8,9,16}. It consists of two major subunit classes, *a* and *b*, with sedimentation coefficients between 30 and 40S. In addition it contains several minor heterogeneous RNAs and possibly several tRNAs¹⁷. Thus the A-rich fraction of 60-70S RSV RNA could be an integral part of its major 30-40S subunits or it could be a distinct species of low molecular weight physically linked to 60-70S RNA. To distinguish between these two alternatives we have carried out the following two experiments.

(1) The presence of an A-rich sequence confers to single-stranded RNA the capability of binding to 'Millipore' filters¹. The 60-70S RSV RNA binds completely and fractionated 30-40S RSV RNA subunits bind almost completely to 'Millipore' filters (Table 3), implying that all subunits of 60-70S RSV RNA contain an A-rich stretch. Not all, however, unfractionated but heat-dissociated 60-70S RSV binds to 'Millipore' filters (Table 3). The fraction that does not bind may represent fragments of RSV RNA lacking an A-rich

Table 3 Binding of RNAs to 'Millipore' Filters

	60-70S RSV RNA*		30-40S RSV RNA*	4-12S RSV RNA†			rRNA‡
	³ H-A	³ H-U	³ H-A	³ H-A	³ H-U	³ H-A	
Native	100 ± 5	100 ± 5	—	13 ± 2	5 ± 2	3 ± 1	—
Heat-denatured	75 ± 5	75 ± 5	95 ± 5‡	10 ± 2	3 ± 2	2 ± 1	—
RNAase-resistant segments	100 ± 5	—	—	100 ± 5	—	—	—

* Isolated after 3-4 h labelling periods.

† Isolated after 8-12 h labelling periods.

‡ Prepared from formaldehyde gradients (Fig. 3).

The numbers represent the percentage of radioactivity of a given RNA sample which is retained by a 'Millipore' filter¹. All values are the average of two to four determination ± range. Between 200 and 2,000 c.p.m. of ³H-RNA were used per experiment. Heat-denaturation was as described previously^{8,9}.

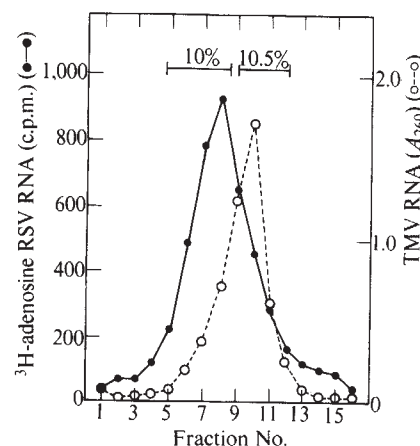


Fig. 3 Distribution of ³H-A-RSV RNA and unlabelled tobacco mosaic virus (TMV) RNA after sedimentation in a sucrose gradient containing 1.1 M formaldehyde. The 60-70S ³H-A RSV RNA was prepared by adding 100 µCi ³H-A to a dense culture of RSV transformed chick embryo fibroblasts. After incubation for 4.5 h the medium was collected. The culture was then incubated for another 3 h after the addition of 50 µCi ³H-A. The medium was collected and the culture once more incubated without further addition of radioactive precursor. The media were then pooled for virus purification and extraction of 60-70S RSV RNA^{8,9}. The ³H-A 60-70S RSV RNA and 100 µg TMV RNA were dissolved in 300 µl. 0.1 M NaCl, 0.02 M EDTA, pH 7.0, 0.1% SDS and 1.1 M formaldehyde and incubated for 15 min at 65° C in a sealed ampoule. Sedimentation was in a 10-25% sucrose gradient containing the same buffer in a Spinco SW 65 rotor at 65,000 r.p.m. for 135 min at 20° C. —, RNAase resistant fraction of RSV RNA.

sequence. Fragmentation of RSV RNA in the virus as a function of time of incubation in the growth medium at 37° C has been described previously^{16,19}. No significant binding was observed with 28S or 18S chick cell ribosomal RNA.

(2) 60-70S ³H-A-RSV RNA was dissociated into subunits in 1.1 M formaldehyde which denatures bihelical regions of nucleic acids¹⁸. The sedimentation distribution of formaldehyde-dissociated RSV RNA relative to that of TMV RNA (Fig. 3) is similar to that of heat or dimethylsulphoxide-dissociated Bryan-RSV RNA described previously⁸. Both the leading and the trailing halves of formaldehyde-dissociated RSV RNA contain the same percentage of RNAase-resistant adenylyl as 60-70S RSV RNA (Table 1, Fig. 3).

We conclude from these two experiments that an A-rich sequence is covalently linked to all 30-40S subunits of RSV RNA. It can be calculated that each 30-40S subunit of 60-70S RSV RNA may contain one or two poly A regions. The molecular weight of the 60-70S RSV RNA has been estimated to be 10-12 × 10⁶ and its adenylic acid content to be about 25%²². Since 9% of the adenylic acid was found to be in an A-rich sequence the total molecular weight of A-rich sequences in 60-70S RNA is about 250,000. Likewise 2-3% of the ³²P-60-70S RSV RNA or that corresponding to a molecular weight of about 250,000 were found to consist of A-rich sequences. If one assumes that 60-70S RSV RNA consists of three or four 30-40S subunits^{8,9,16}, it follows that each subunit may contain one or two A-rich sequences of a molecular weight of 60,000.

The RNAs of tumour viruses of all known taxonomic groups have many properties in common^{16,22}. We have therefore analysed the 60-70S RNA of Rauscher mouse leukaemia virus (MLV) in the same way as RSV RNA to test for the presence of an A-rich sequence. About 10(±0.5)% of the radioactivity of ³H-A 60-70S MLV RNA and about 1(±0.2)% of the radioactivity of ³H-U 60-70S MLV RNA were RNAase-resistant in the conditions described for Table 1. All 60-70S MLV RNA was found to bind to 'Millipore' filters, and the A-rich fraction of MLV RNA has a sedimentation coefficient of about 4.5S (Fig. 1b). We conclude that 60-70S

MLV RNA contains an A-rich sequence very similar to that of RSV RNA.

Analysis of the RNA of the cytotoxic influenza virus gave the following results. About $2(\pm 0.5)\%$ of the radioactivity of $^3\text{H-A}$ and about $1(\pm 0.2)\%$ of the radioactivity of $^3\text{H-U}$ influenza virus RNA were resistant to RNAase. The sedimentation distribution of the RNAase-resistant fraction of $^3\text{H-A}$ influenza virus RNA was heterogeneous. Three ($\pm 1\%$) of the radioactivity of $^3\text{H-A}$ or $^3\text{H-U}$ influenza virus RNA was found to bind to 'Millipore' filters. We conclude that influenza virus RNA does not contain an A-rich sequence similar to that found in the RNAs of RNA tumour viruses.

The size and homogeneity of the A-rich sequences of RSV RNA or MLV RNA are very similar to those of poly A regions in the mRNAs of animal cells^{1,2}. If poly A sequences are involved in transport of mRNA from the nucleus to the cytoplasm¹⁻³ it may be speculated that tumour virus RNA is replicated in the nucleus and that an A-rich sequence is attached to transport it to the cytoplasm. This speculation is compatible with other evidence suggesting that the RNA of RNA tumour viruses may be replicated in the nucleus^{20,21}.

The capacity of RSV RNA to bind to 'Millipore' filters may be useful in detecting tumour virus RNA in preparations of virus-infected cells. Further experiments will be necessary to determine whether A-rich sequences are involved in initiation of DNA synthesis by the tumour virus DNA polymerase and whether they may be involved in linking the subunits of 60-70S tumour virus RNA together. The presence of poly A sequences in tumour virus RNA probably has to be considered in interpreting the various hybridization experiments of cellular nucleic acids with tumour virus RNA or DNA transcribed from RNA with the viral DNA polymerase. Drs M. Green and M. Cartas (St Louis) have informed us that they have also found a poly A sequence in murine and avian tumour virus RNA.

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New Possibilities of Investigating Nucleic Acids and Nucleoproteins in Aqueous Solutions by Infrared Spectroscopy

A NEW approach to the study of nucleic acid structure lies in the investigation of infrared spectra in the 950-1,350 cm^{-1} range where the main bands represent the sugar-phosphate backbone vibrations¹. In principle, water transmittance in this region enables spectra of aqueous solutions to be obtained² and the absence of strong protein bands provides a unique opportunity for investigating nucleic acid structure inside the nucleoprotein particles. We have carried out temperature-dependent measurements of DNA and RNA spectra both in the free state and within viruses. Our results show that the spectra reflect accurately the peculiarities of the sugar-phosphate skeleton of nucleic acids.

The total calf thymus DNA and yeast RNA (Sigma), phage SD and tobacco mosaic virus (TMV) were investigated. The concentration as calculated for nucleic acids was 1-3%. Special demountable cells with a layer thickness of 40 μm , with one window of 'Irtran' glass and the other of calcium fluoride, were used. Spectra were recorded by the Perkin-Elmer 180 spectrophotometer using a double chopping system which eliminates the sample's own radiation.

Many of the absorption bands of native DNA (Fig. 1) can be assigned on the basis of earlier work¹, yet the band at

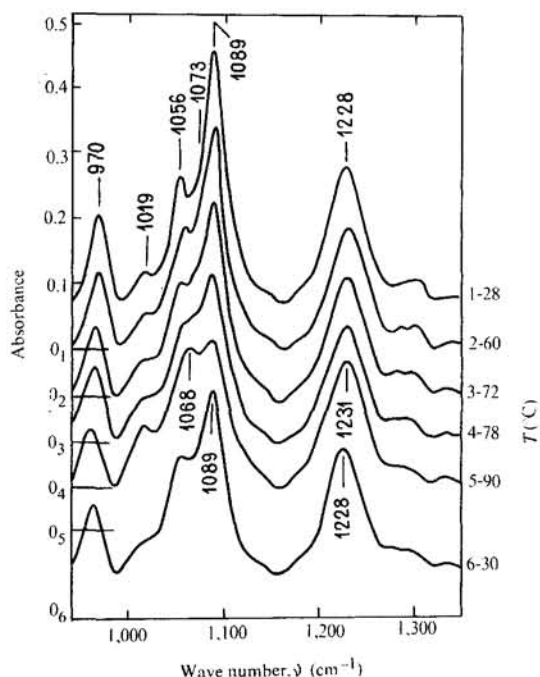


Fig. 1 Infrared spectra of calf thymus DNA in aqueous solution at different temperatures. Concentration 2.7% in 0.014 M NaCl, 0.001 M $\text{CH}_3\text{COONH}_4$ (pH 7). Absorption curve numbers and solution temperature are given on the right. The zero line of each absorption curve has the index corresponding to the curve number.

$1,056\text{ cm}^{-1}$, the most inherent to native DNA, has no exact assignment; it can be attributed either to the skeleton stretching vibration of the $\text{C}_5\text{-O}_4$ ester bond or to the sugar ring vibration. On the thermal denaturation it is substituted by a new band at $1,068\text{ cm}^{-1}$. Similar changes in the position of this band were observed when the DNA was denatured by other methods². The maximum of one of the phosphate bands increases smoothly from $1,228\text{ cm}^{-1}$ to $1,231\text{ cm}^{-1}$. This is probably due to some weakening of the hydrogen bonding between the phosphate group and water molecules. The spectrum of the renatured DNA after cooling to 30°C is very similar to that of the initial form.

The spectrum of phage SD, lysing *E. coli* SK³ and containing 46% DNA, does not differ significantly from that of the free nucleic acid (Fig. 2). The equality of the frequencies of the phosphate bands of intraphage and free DNA in solution and in films⁴ indicates that PO_4^- has the same water environment. The peak of $1,056\text{ cm}^{-1}$ has the same intensity as in the free native molecule. Heating the phage suspension to 61°C disrupts the phage coat and releases the DNA into the solution⁵ without spectral changes. Cooling to 30°C leads to almost complete reconstitution of the spectrum. Thus we can assume that the sugar-phosphate skeleton structure of the intraphage DNA is nearly the same as that of the native free state. The possibility of conformational changes in other parts of the DNA inside the phage is not excluded⁶.

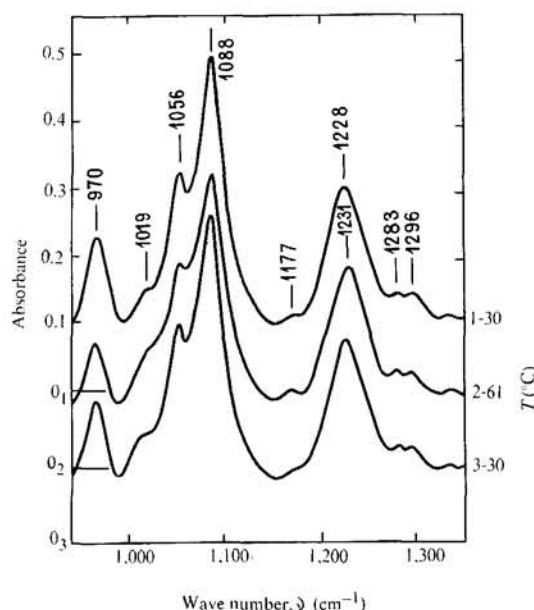


Fig. 2 Infrared spectra of phage SD suspension in aqueous solution at different temperatures. Concentration calculated for DNA 3% in 0.14 M NaCl , $0.001\text{ M CH}_3\text{COONH}_4$ ($\text{pH } 7$). Designations are the same as in Fig. 1.

The yeast RNA spectrum (Fig. 3) differs considerably from that of DNA. The characteristic feature is the splitting of the phosphate band near $1,230\text{ cm}^{-1}$ with $\Delta\nu=18\text{ cm}^{-1}$. This may be attributed to the interaction of the phosphate vibrations along the ordered helical chain⁷. Another explanation is the presence of internal binding of the phosphate groups with other molecule groups, for example, hydrogen bonding with 2'-hydroxyl of ribose which can stabilize the secondary structure of RNA⁸. Heating and denaturation of RNA evoke considerable changes in the spectrum: all the bands are weakened and broadened; the 994 cm^{-1} band disappears and the splitting of the phosphate band decreases smoothly until a single band appears at $1,230\text{ cm}^{-1}$ of the lesser peak intensity. Cooling of the solution leads to reconstitution of the initial spectrum. The splitting of the phosphate band is now 15 cm^{-1} , which corresponds to the renaturation of RNA.

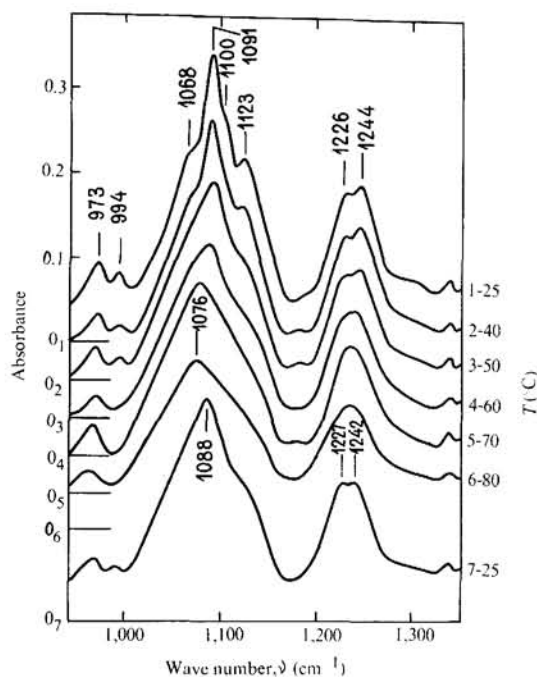


Fig. 3 Infrared spectra of yeast RNA in aqueous solution at different temperatures. Concentration 2.7% in 0.014 M NaCl , $0.001\text{ M CH}_3\text{COONH}_4$ ($\text{pH } 7$). Designations are the same as in Fig. 1.

The RNA in the TMV is only about 5% of its dry weight and its spectrum differs from that of free RNA (Fig. 4). The amide III band of protein has a maximum near $1,300\text{ cm}^{-1}$. Assuming that the antisymmetric phosphate band is split into $1,249$ and $1,213\text{ cm}^{-1}$, $\Delta\nu$ has the large value of 36 cm^{-1} . The $1,068\text{ cm}^{-1}$ band of free RNA is not observed in the TMV spectrum. The data confirm that the intravirus RNA has a special structure⁹. Heating the sample above 60°C causes disruption of the virus protein coat, which has stabilized the structure of RNA, and the release of RNA into solution¹⁰. This causes a decrease of the amide band, and the nucleic acid becomes denatured with a spectrum very similar to that of the free RNA, in particular the phosphate band splitting is cancelled. Cooling the suspension causes a further change in the spectrum: the phosphate band again splits into bands at

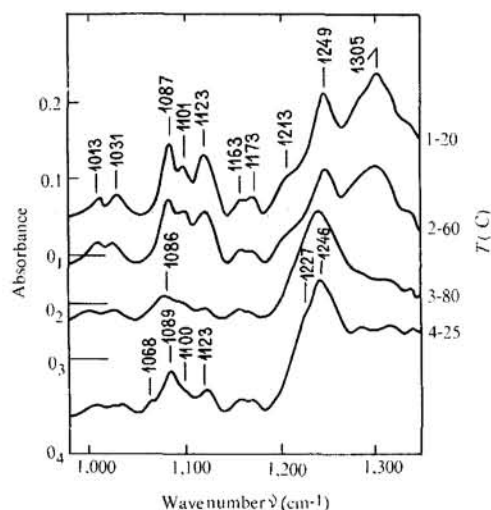


Fig. 4 Infrared spectra of tobacco mosaic virus in aqueous solution at different temperatures. Concentration 20% (1% of RNA) in 0.014 M NaCl , $0.001\text{ M CH}_3\text{COONH}_4$ ($\text{pH } 7$). Designations are the same as in Fig. 1.

1,227 and 1,246 cm^{-1} and the 1,068 cm^{-1} band of free RNA appears. In general it follows from the spectrum that the state of TMV RNA now corresponds to that of the free molecule with double-stranded regions of the secondary structure.

Thus the application of the infrared method for studying nucleic acid both in the free state and within the nucleoprotein complexes in water solution at this time can give new important information about the structure. A detailed assignment of bands and the use of quantitative criteria will serve to develop the method further.

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Reaction between Molecular Oxygen and Photo-excited Protoporphyrin IX

THE absorption spectrum of protoporphyrin IX in tetrahydrofuran solution has maxima at 404 (Soret band), 503, 536, 576 and 633 nm. Excitation at any of these wavelengths produced an intense red emission with its maximum at 635 nm¹. The same emission spectrum was observed when added biacetyl was irradiated at its 436 nm absorption. In these conditions the biacetyl phosphorescence was completely quenched. Additionally, molecular oxygen quenched the protoporphyrin IX emission spectrum following Stern-Volmer kinetics. Typical experimental results, obtained at 25° C, are plotted in Fig. 1.

Hence the emitting state of the porphyrin can be sensitized by triplet biacetyl and quenched by (triplet) molecular oxygen. This strongly suggests that the emitting state is itself a triplet²⁻⁴. It has previously been considered to be a singlet¹, and its further characterization is in progress.

Transfer of excitation energy from the triplet state of an organic sensitizer molecule to normal molecular oxygen ($^3\Sigma_g^-$) leaves the oxygen molecule in a singlet metastable state ($^1\Delta_g$ or $^1\Sigma_g^+$)^{5,6}. Thus a metastable state of the oxygen molecule should be formed in the quenching process.

The type II photo-oxidation of furans⁷ has been used as a diagnostic test for the presence of $^1\Delta_g$ oxygen⁸. We have identified the principal product of protoporphyrin IX sensitized photo-oxidation of furan in methanol at -78° C. Its infrared, nuclear magnetic resonance, and low resolution mass spectra were consistent with the known⁷ product of type II photo-oxidation. This product was not detected in the absence of

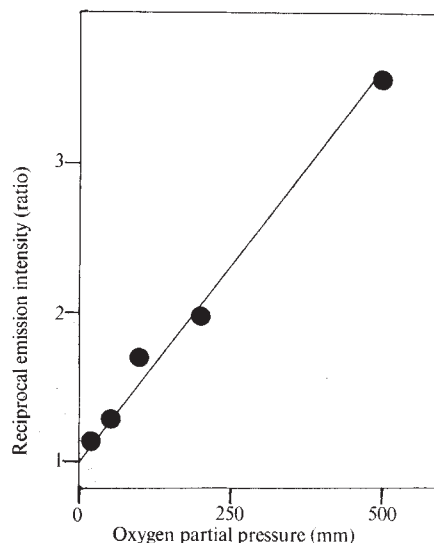


Fig. 1 Stern-Volmer plot for solution of protoporphyrin IX in dry degassed tetrahydrofuran under oxygen. The results remained reproducible after several cycles of freeze-thawing under high vacuum.

porphyrin. We regard this as chemical evidence for the presence of $^1\Delta_g$ oxygen in the reaction mixture.

This chain of observations clearly implies a mechanism for the generation of $^1\Delta_g$ oxygen in porphyrin containing biological systems. The presence of free protoporphyrin in surface tissue is characteristic of some types of the human disease porphyria⁹, which are characterized by extreme sensitivity to sunlight. Our results implicate $^1\Delta_g$ oxygen in the mechanism of photo-damage and suggest that therapy by competitive quencher molecules, such as β carotene, might be effective in such cases.

It was recently reported independently that clinical trials of β carotene therapy were in progress¹⁰.

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Couplings of Enzymes onto Microorganisms

THE coupling of enzymes onto insoluble support materials offers several advantages including increased stabilization of the enzyme, re-use of the enzyme without tedious recovery by precipitation, and close control of the extent of conversion in enzyme reactors. Coupling may also afford an improved heat-stability, greater specificity, or a different pH optimum compared with the enzyme in the free state. On the whole, work on insolubilized enzymes has been confined to single enzymes separated from the organism that produced them and coupled to non-living supports¹⁻⁴. The work reported here concerns the coupling of enzymes onto living cells in order to supplement the array of enzymes naturally produced.

It is well known that yeast cells secrete a number of enzymes including invertase, phosphatase and proteinase that become coupled naturally either by entrapment or covalent linkage to the mannan of the cell wall⁵. If cereal flour is added to a medium in which yeast is growing, β -amylase present in the flour and not native to the yeast is readily adsorbed onto the surface of the yeast cell walls⁶. In this case, the adsorption is insufficiently strong to prevent some of the enzyme being washed off subsequently and covalent linkage would therefore be preferred.

Insolubilization of enzymes by covalent linking has involved methods that are rather drastic for living organisms so that it is highly unlikely that they would remain viable if used as supports. A technique has been developed by Novais for covalent coupling using titanium salts⁷. Although in its original form it is too harsh, a modification provides more gentle treatment. Principally, the modifications have entailed lowering the concentration of the titanium salts, raising the pH levels during treatment and avoiding steps where the cells would desiccate.

In a typical treatment, 1.2 g of washed pressed brewery yeast with no amyloglucosidase (glucamylase) activity was stirred for 15 min at ambient temperature in titanic chloride (0.125% w/v) and then washed thrice in sodium acetate buffer at pH 4.5. It was held overnight at 4° C in the presence of amyloglucosidase before repeated washings in the buffer and saline. The enzyme was covalently bound because the washings failed to bring it into solution and because the yeast displayed amyloglucosidase activity.

The activity of the yeast was measured as mg glucose released per min by 1 g dry weight of yeast from 1% Merck soluble starch in 0.005 M sodium acetate buffer at pH 4.5, the assay being carried out at 45° C. The glucose was measured after removal of yeast and free amino-acids by the Nelson-Somogyi method. Typically, activities were in the range 5.3–8.3 after due allowance for blank values and the viable counts were in the range 50–80% as indicated by slide-culture procedures⁸. This is illustrated in Table 1.

Table 1 Effect of Using Titanic Chloride for coupling Amyloglucosidase to Brewers' Yeast

Treatment	pH	% Viability (slide culture)	Enzyme activity (mg glucose g ⁻¹ dry wt yeast min ⁻¹)
None	4.5	95	0
Enzyme preparation in water only	4.5	60	0
2.5% TiCl ₄ added only	1.0	10	0
0.125% TiCl ₄ added only	2.2	55	0
2.5% TiCl ₄ and enzyme used	1.0	10	17.1
0.125% TiCl ₄ and enzyme used	2.2	60	1.5
2.0% TiCl ₃ and enzyme used	1.6	30	4.0

Treatment consisted of three washings in 0.005 M acetate buffer (pH 4.5), stirred at 4° C with enzyme solution overnight (activity of the solution is 5.43 mg glucose mg⁻¹ protein min⁻¹) and then washed four times alternately in acetate buffer and in acetate buffer plus M sodium chloride, followed by four additional washings in acetate buffer. Results represent mean of three estimations.

A range of inorganic salts can be used as coupling agents with varying degrees of efficiency, including titanous, stannous and ferrous salts. Several strains of *Saccharomyces cerevisiae* have been employed successfully including strains 1004, 1005, 1026 and 1040 of the National Collection of Yeast Cultures, Brewing Industry Research Foundation, Nutfield, Surrey. A range of enzymes including bacterial α -amylase and trypsin have been coupled. Furthermore, amyloglucosidase has been successfully coupled on to *Bacillus subtilis* and *Escherichia coli*.

The coupling of amyloglucosidase to yeast permits hydrolysis of dextrans to yield simple sugars that the yeast can assimilate and ferment. Such a procedure would have value in the production of beer, vinegar, whisky or industrial ethanol. Other possible uses for enzymes coupled to yeast include the breakdown of protein in a medium if proteinase is coupled. This means that hazes involving protein⁹ could be removed by yeast. At the same time, the presence of yeast would maintain a low redox potential that is valuable in beverages such as beer. The simple treatment of yeast or bacteria as described could be used to augment their array of hydrolytic enzymes and provide a means of degrading a wider range of components in complex media. It also affords the opportunity of removing enzymes at a selected juncture, merely by centrifuging, filtering or precipitating the microorganisms.

When microorganisms that have been coupled multiply, the average enzyme activity of the individual cells will fall because of denaturation and because the new cells will not receive the coupled enzyme. In closed continuous fermenters, however, cell multiplication is low¹⁰ and therefore the overall enzyme activity is likely to be maintained. The technique of coupling enzymes to living supports is in its infancy but could well be of wide application. It is envisaged that comparatively inexpensive enzymes will be used in many applications. The coupling procedure is simple, cheap and rapid so that the microorganisms can be coupled with fresh enzyme as required.

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Platelets and Experimental Scurvy

HAEMORRHAGE in scurvy has been attributed to increased capillary fragility caused by reduced collagen formation¹ and consequent changes in vascular endothelium and perivascular connective tissue²⁻⁴. Platelet adhesiveness to glass is impaired in human scurvy⁵⁻⁷ and in experimental scurvy in guinea-pigs^{8,9}. Born and Wright¹⁰ have reported a diminished velocity of adenosine diphosphate (ADP)-induced aggregation of platelets from scorbutic animals. Platelet adherence to exposed subintimal collagen, release of platelet ADP, and subsequent platelet aggregation are believed to be early events in the formation of a haemostatic plug¹¹. These observations suggest that a platelet defect may contribute to scorbutic haemorrhage. We report here results of experiments which

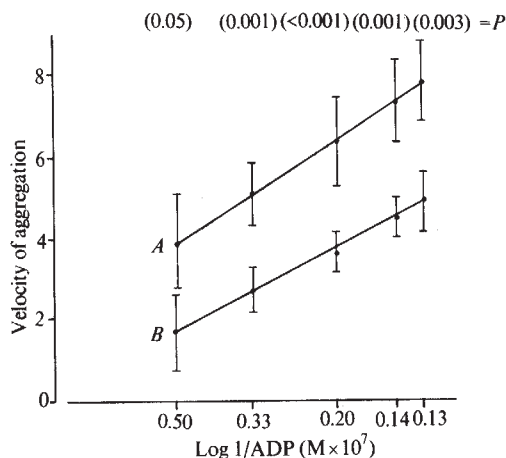


Fig. 1 Velocity of aggregation of normal and scorbutic guinea-pig platelets caused by ADP. Each value is the mean $\pm$ s.d. of four to seven observations at each ADP concentration in at least three pools of platelet-rich plasma. A, Normal platelets; B, scorbutic platelets. In this and succeeding figures, numbers in parentheses are *P* values calculated by Wilcoxon's rank sum test.

confirm the work of Born and Wright¹⁰ with respect to diminished velocity of ADP-induced aggregation of scorbutic guinea-pig platelets *in vitro*. In addition, we have investigated the interaction of collagen and platelets from both normal and scorbutic animals.

In each of five experiments, groups of four to twenty-four male Hartley guinea-pigs were fed water and either standard ascorbic acid-supplemented guinea-pig chow (Ralston Purina Co.) or ascorbic acid-deficient chow (Nutritional Biochemical Corp.) *ad lib*. Within 3–4 weeks, animals on the ascorbic acid-deficient diet developed symptoms characteristic of scurvy: loss of hair, petechial haemorrhages, and a mean net loss in body weight of 89 g (± 39 g, s.d.). Animals given the standard diet were healthy and normal in appearance and had gained weight (151 ± 38 g).

As soon as scorbutic symptoms were recognized, platelet-rich plasma was separated from citrated whole blood of normal and scorbutic guinea-pigs as described for the rabbit¹². Platelet-rich plasma from two to four animals was pooled. Platelets were counted in whole blood and in platelet-rich plasma¹³. Collagen was prepared as a crude extract of freshly excised normal or scorbutic guinea-pig hind leg tendon, as described for rabbit Achilles tendon¹⁴, and is referred to as "tendon extract" (TE). Platelet aggregation caused by the addition of ADP or TE to platelet-rich plasma was studied at room temperature by a modification¹² of Born's turbidimetric method¹⁵. Aggregation velocity was calculated as the initial maximum rate of change in light transmittance¹⁶, and was divided by the number of platelets ml^{-1} platelet-rich plasma to permit comparison of different pools of plasma.

In each experiment, the platelet counts of scorbutic guinea-pigs were higher than those of normal animals, and this difference was significant when the mean counts were compared (Table 1). Barkhan and Howard¹⁷ reported similar results,

but Born and Wright⁸ did not observe a significant change in platelet count in their experiments. Normal or low platelet counts have been reported in human scurvy¹⁸; the significance of this apparent species difference is not known.

The velocity of aggregation of normal guinea-pig platelets caused by ADP was significantly greater than that of scorbutic platelets at each concentration of ADP tested (Fig. 1), which confirms the work of Born and Wright¹⁰. Guinea-pig platelets aggregate biphasically in response to low concentrations of ADP¹⁹, and the second phase of aggregation is associated with release of platelet ADP²⁰. The incidence of biphasic responses was approximately the same in normal and scorbutic platelet-rich plasma at concentrations of $2\text{--}5 \times 10^{-7}$ M ADP, suggesting that the platelet defect in experimental scurvy is probably not related to a gross defect in platelet ADP release.

Platelets from scorbutic animals aggregated following addition of either normal or scorbutic TE to platelet-rich plasma. Because there is strong evidence that TE-induced platelet aggregation is mediated by the release of platelet ADP^{21,22}, this supports our suggestion that scorbutic platelets do not have a functional defect in ADP release. The velocity of aggregation of scorbutic platelets was significantly lower than that of normal platelets at each dilution of normal or scorbutic TE tested (Figs. 2 and 3). The platelet defect in scurvy is thus apparently related to the platelet response to ADP, whether released from platelets following their adhesion to collagen fibres or added exogenously, and not to a defect in platelet ADP release.

Although the aggregation velocity of scorbutic platelets caused by scorbutic TE (Fig. 3B) was slightly lower than that caused by normal TE (Fig. 2B), the difference was not significant. Similarly, the aggregation velocity of normal platelets caused by scorbutic TE (Fig. 3A) was not significantly different from that caused by normal TE (Fig. 2A). Collagen is most likely the aggregating agent in TE^{22–24}, so these results suggest that scorbutic collagen does not contribute to the diminished velocity of collagen-induced platelet aggregation in scurvy. This is consistent with the observations that although total collagen synthesis is reduced in scurvy¹, the amino-acid composition is the same as in collagen from normal animals²⁵. The free amino groups required for the interaction of collagen with platelets²⁴ would therefore be present in the same proportion in both scorbutic and normal collagen.

Although we have confirmed the diminished velocity of ADP-induced aggregation of scorbutic platelets¹⁰, and have observed the same phenomenon during TE-induced aggregation, we doubt this implies impaired platelet function in scorbutic haemorrhage. Harrison and Honour⁹ observed that

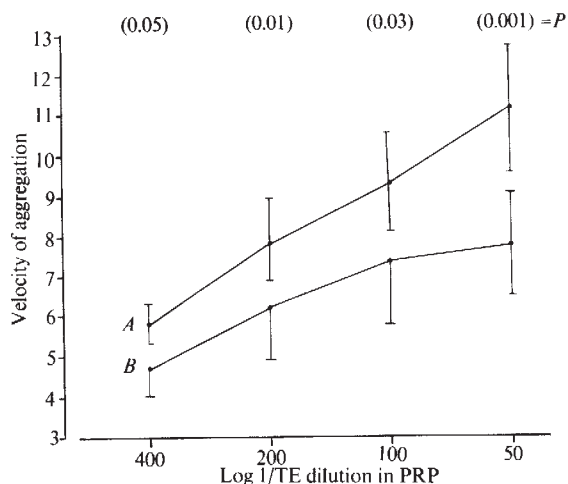


Fig. 2 Velocity of aggregation of normal and scorbutic guinea-pig platelets caused by TE from normal animals. Each value is the mean $\pm$ s.d. of four to ten observations at each TE dilution in at least three pools of platelet-rich plasma. A, Normal platelets; B, scorbutic platelets.

Table 1 Whole Blood Platelet Counts of Normal and Scorbutic Guinea-pigs

Experiment	Platelets mm^{-3} (mean $\pm$ s.d. $\times 10^{-5}$)	
	Normal guinea-pigs	Scorbutic guinea-pigs
I	6.85 ± 1.58	8.29 ± 2.49
II	5.25 ± 0.83	8.95 ± 1.41
III	6.41 ± 1.31	7.86 ± 1.30
IV	7.84 ± 0.93	10.24 ± 1.32
V	5.06 ± 0.63	7.98 ± 1.00
Mean $\pm$ s.d.	$*6.28 \pm 1.15$	$*8.66 \pm 0.98$

* $P < 0.01$ (Student's *t* test).

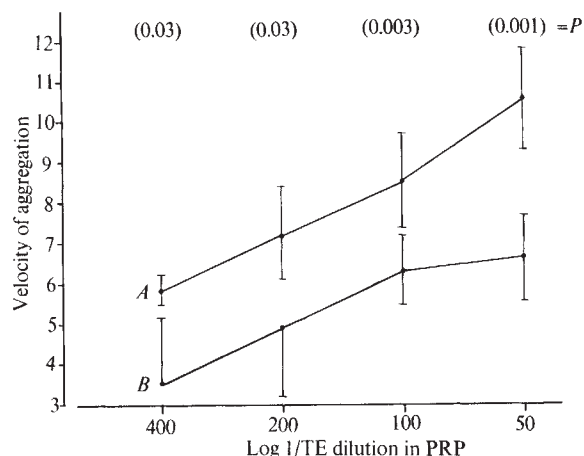


Fig. 3 Velocity of aggregation of normal and scorbutic guinea-pig platelets caused by TE from scorbutic animals. Each value is the mean $\pm$ s.d. of four to twelve observations at each TE dilution in at least three pools of platelet-rich plasma. A, Normal platelets; B, scorbutic platelets.

haemostatic plug formation in scorbutic guinea-pigs is not impaired, and concluded that haemorrhage could not be attributed to failure of platelets to adhere and form effective plugs. Our results support this, for we present evidence that there is no defect in *in vitro* adhesion of scorbutic platelets to collagen or in release of platelet ADF. Haemorrhage in scurvy is chiefly from capillaries and is probably due to defects in endothelial structure caused by depletion of the supporting perivascular collagen⁴, although a decreased synthesis of sulphated mucopolysaccharides may also be implicated^{3,25-28}. Platelets may have some role in maintaining normal capillary permeability^{8,9}, but the *in vitro* platelet defects described so far—impaired adhesiveness to glass and diminished velocity of aggregation—do not seem to be related to the increased capillary fragility observed in scurvy.

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Changes during Maturation and Metamorphosis in the Synaptic Organization of the Tadpole Retina Inner Plexiform Layer

ELECTROPHYSIOLOGICAL studies have shown that there is considerable variation in the degree of processing of visual information carried out by the retinas of different species, apparently correlated with anatomical difference between species. In amphibian and avian retinas with complex receptive fields there seem to be more conventional (amacrine cell) synapses than in mammalian retinas with simple organization. Moreover, the greater density of serial-conventional synapses, in which a presynaptic amacrine process is also postsynaptic to another amacrine process, is particularly striking in complex retinas. The density of ribbon (bipolar cell) synapses remains fairly constant from species to species¹. The fact that tadpoles apparently make little use of visual cues, whereas adult frogs depend on them for survival, suggests that electrophysiological and anatomical retinal reorganization occurs during metamorphosis. There is some evidence for this^{2,3}. Here I report changes in retinal synaptic organization found in *Rana pipiens* both during maturation and metamorphosis and as a result of a hormone pellet implantation.

Electron micrographs of the inner plexiform layer at a magnification of 25,000 times were used to construct mosaics covering the area from the ganglion cell layer to the vitreal margin of the inner nuclear layer for eleven separate Taylor-Kollros⁴ larval stages and the adult frog. There are three unambiguously recognizable types of synapses in the inner plexiform layer^{5,6}: ribbon synapses, found only in axon terminals of bipolar cells; conventional synapses, found in amacrine cell processes; and serial-conventional synapses—amacrine to amacrine connexions. All synapses in a mosaic were classified and counted following the criteria outlined by Dubin¹. The total number of each synaptic type was divided by the area of the mosaic to obtain the density of synapses per unit area of inner plexiform layer per section. Because the retina increases in thickness (from vitreal to scleral margins) from Taylor-Kollros stage I to adulthood, the difference in densities from stage to stage is not directly proportional to the difference in number of synapses per radial column of retina between those stages. To determine the relative number of synapses at each larval stage, each density calculated was multiplied by the ratio of the thickness of the inner plexiform layer of that stage to that of the adult. Measurements made at several stages suggest that the inner plexiform layer thickness increases linearly with larval stage. This linear approximation was used to calculate the values of adjusted densities.

Adjusted density of conventional synapses is plotted as a function of larval stage (upper curve, Fig. 1A): the positive slope indicates that conventional synapses are constantly being added. Growth of the inner plexiform layer is therefore not simply an increase in number of processes but is accompanied by the formation of new amacrine synapses. Some increase in the rate of addition of conventional synapses with stage is apparent from stages XVI to XX. Ribbon synapses too are being added during larval life (lower curve, Fig. 1A) and

maintain a constant rate of addition with the stage of development until adulthood. In striking contrast to the regular increase in ribbon synapses is the rapid addition of serial-conventional synapses from stages XVI to XX (Fig. 1B). The late larval stages and the adult are characterized by an adjusted density of serial-conventional synapses almost six times that of the stages just before stage XVI. This sudden increase in synaptic density is correlated in time of development with the onset of metamorphosis.

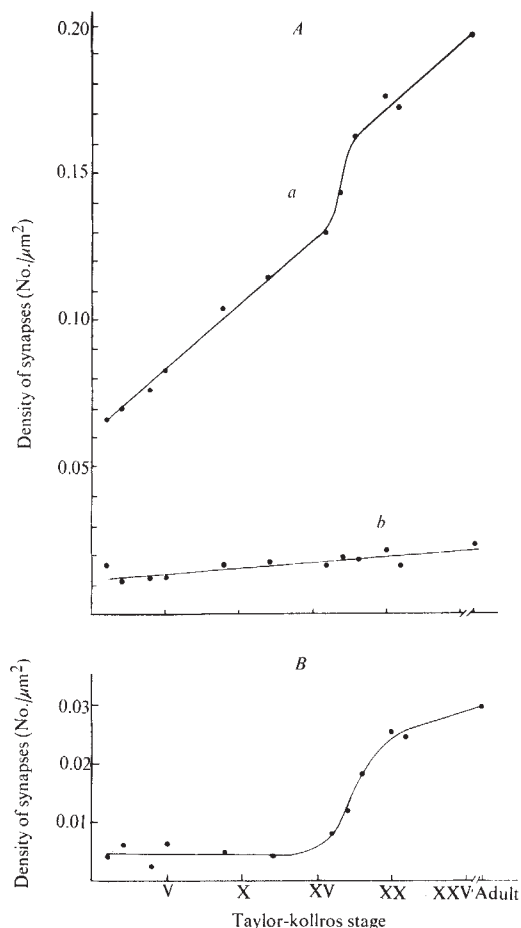


Fig. 1 A, Densities of synapses adjusted for a linear increase in width of inner plexiform layer from stage I to adulthood plotted as a function of Taylor-Kollros stage. a, Adjusted conventional synapse density; b, adjusted ribbon synapse density. B, Adjusted density of serial-conventional (short distance amacrine to amacrine) synapses versus Taylor-Kollros stage. A location near the optic nerve was chosen in order to have a reference point for sampling approximately the same area from retina to retina. Each point represents mosaics covering approximately 1,000 μm^2 of inner plexiform layer. There were, on the average, 200 conventional, thirty ribbon and fifteen serial-conventional synapses per mosaic. In all, nearly 3,000 synapses were classified to yield these curves.

The stages between XV and XX are characterized by various morphological and biochemical changes, many of which have been shown to be triggered by thyroxine^{7,8}. To determine if the dramatic increase in serial-conventional synapse density can be induced by thyroxine, 100 μg pellets of 10% thyroxine and 90% cholesterol were implanted in the vitreous body of one eye of stage VII tadpoles. Pellets of cholesterol without thyroxine were implanted in the eyes of control tadpoles and tadpoles, untreated except for the MS-222 anaesthetic as used on the operated animals, were raised as normal controls. Both the cholesterol and normal controls progressed from stage VII to IX over 116 h of the experiment. Stage IX occurs well

before the onset of metamorphosis or the normal increase in serial-conventional synaptic density of stage XVI.

In all cases the density of the serial-conventional synapses of the treated eye (T) was greater than that of the untreated eye (U), and those of the normal and cholesterol controls. It is clear that the response of the tissue had started sometime before 67 h with the maximal effect seen at about 92 h. The densities for the treated and untreated eyes were separately averaged and compared. Average density of the serial-conventional synapses in the treated eye increased over that of the untreated eye by 64.3% and over the average of the controls by 92%. The same comparisons for ribbon and conventional synapses are: ribbon, -18.5% from untreated and -8.3% from the control eye; conventional, +3.8% from the untreated and +9.2% from the control eye. Thus thyroxine causes a precocious increase in density of serial-conventional synapses while the ribbon synapses remain unaffected by the hormone.

Table 1 Density of Ribbon (R), Conventional (C) and Serial-conventional (S) Synapses in Thyroxine Treated (T) and Untreated (U) Eyes from the Same Animal compared with Controls

Eye	Density of synapses (No./1,000 μm^2)		
	R	C	S
T (average of four eyes)	22	190	23
U (average of four eyes)	27	183	14
Cholesterol control	25	176	14
Stage IX control	23	172	9

It is not possible, from this study, to determine if the increase in numbers of serial-conventional synapses is due to the appearance of new amacrine cells, or to the proliferation of new processes which then establish connexions with other cells. Hollyfield⁹ has reported that cells that have divided in the peripheral retina of *R. pipiens* tadpoles migrate through the inner nuclear layer and take up their final positions in the central portion of the retina, but the identity of these cells, and whether or not they are neural, is unknown. Of primary interest is the functional significance being subserved by this anatomical reorganization. Reuter² has reported a possible change in class 2 ganglion cell fibres between tadpole and adult in a related species, while Pomeranz and Chung³ have described the premetamorphic tadpole retina as having no class 1 ganglion cell fibres and few class 2 fibres at the periphery. Both types of fibre are associated with moving stimuli¹⁰, which suggests a functional relationship between the anatomical changes and the reported changes in motion sensitivity. Furthermore, directionally selective cells have been reported in the retina of the adult frog¹¹ but not in the tadpole. Further electrophysiological studies on the tadpole retina might reveal the function of the serial-conventional synapses of the amacrine cells in visual processing.

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I-Leucopenic Effect of *Rhazya stricta*

Two alkaloids obtained from *Vinca rosea*, vinblastine and vincristine, have antileukaemic and anticancer activity and so we have screened other plants of the same family (apocynacea) for similar activity. *Rhazya stricta* Diceisne, a gregarious dwarf shrub which grows wild in West Pakistan and is rich in alkaloids¹⁻⁴, proved to have a leucopenic effect.

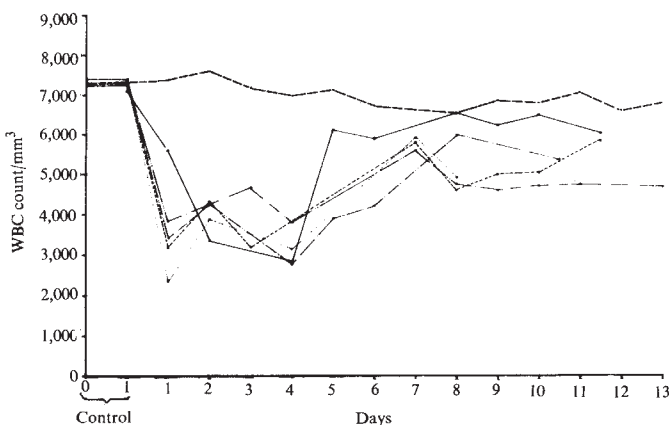


Fig. 1 Leucopenia produced in rats, after a dose of 15 mg/kg body weight. ---, Control.

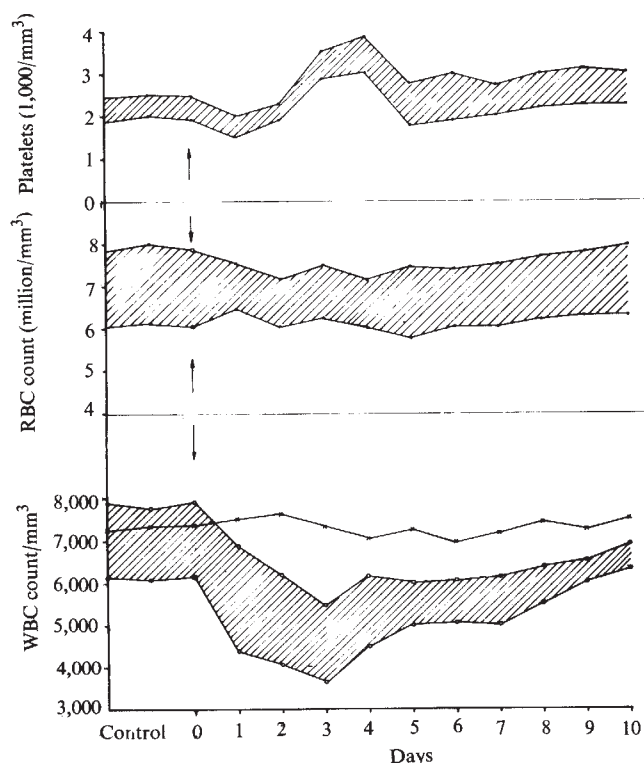


Fig. 2 WBC, RBC and platelet count in albino rats after a single intraperitoneal injection of the *Rhazya stricta* extract.

A single intraperitoneal injection of an extract of *R. stricta* (15 mg/kg body weight) reduced significantly the total white blood cell count of adult albino rats. As Fig. 1 shows, there was a sharp decline on the day after injection and a 50–60% reduction within 3 days. The count returned to normal in 7–10 days. There was no change in the cell count of control rats.

In another group the drug caused a sharp decline in WBC count but did not affect the red cell count, while the platelet count increased slightly. Leucocyte count and platelet count later returned to normal. There was no significant change in the haemoglobin content.

When rats were given 20 mg/kg of the extract orally, there was a marked leucopenic effect. The decrease in the WBC was gradual and lasted 9–10 days (Fig. 2). No rebound leucocytosis was observed.

The rapid onset of leucopenia suggests that extract of *Rhazya stricta* has a selective action on circulating white blood cells. On the other hand, the long duration of leucopenia may be due to a reversible depression of bone marrow. Perhaps the bone marrow is also depressed selectively, and while the WBC count is depressed the RBC count and the haemoglobin level remain normal.

In dogs, an intravenous dose of 80 mg/kg of the extract produced intense salivation and rigor, followed by respiratory failure, convulsions and death within 15 min. Oral subacute toxicity investigation in rats given 150 mg/kg of the drug once a week for 12 weeks showed a loss of about 10% of original body weight. Three out of twenty-five rats died, while two from the control group died from secondary infection. This drug has no side effects on the other systems of the body and its toxicity is also negligible when administered orally. We are now investigating the action of this drug in experimental leukaemia and in animals with transplanted tumours.

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Evolution of Mixed Populations of Genetically Different Mitochondria in *Paramecium aurelia*

MITOCHONDRIA are clearly not strictly autonomous organelles and depend for their biogenesis on both nuclear and mitochondrial information¹⁻⁵. But how autonomous is the mitochondrion with respect to other mitochondria within a cell?

We have tried to answer this question in *Paramecium aurelia* using the method developed for yeast⁶⁻⁹. Two genetically different types of mitochondria are brought together within the same cell and the behaviour and evolution of such "hetero-mitochondriotes" are studied through vegetative growth. A number of cytoplasmically inherited mutations of resistance to antibacterial antibiotics such as erythromycin and chloramphenicol have been obtained in *P. aurelia* (refs. 10 and 11 and unpublished work of A. A. and J. B.). These mutations, E^R or C^R, which are probably located in mitochondrial

DNA^{11,12}, enable *Paramecium* cells to grow normally in the presence of a concentration of antibiotic which completely blocks growth of wild type cells (E^S , C^S).

Heteromitochondriotes are obtained by taking advantage of some properties of *Paramecium*^{13,14} (Fig. 1). Starting either from an exconjugant of pairs that have undergone cytoplasmic exchange, or from "doublets", cells containing mixed cytoplasm are first allowed to divide once in non-selective medium; one cell is placed in selective medium and the other is kept in non-selective medium, yielding a clone which is subcloned every ten fissions. Samples of cells can then be tested when required. Selective medium is the usual bacterized grass infusion¹⁴ +100 g ml.⁻¹ erythromycin or 200 g ml. chloramphenicol. Growth at "high" temperature, 36° C compared with 26° C, also constitutes in some cases a selective condition, for several E^R mutations confer thermosensitivity on the cell. For each test of a clone, three to thirty cells are transferred to the chosen selective condition and the phenotype is determined by recording daily the growth rate and general appearance of the cells. Resistant cells grow at a normal rate, while sensitive cells are blocked after one or two residual fissions and eventually die, and a range of intermediary phenotypes, corresponding to cells that turn progressively resistant after variable lags, is readily distinguishable. The list of the combinations studied is given in Table 1.

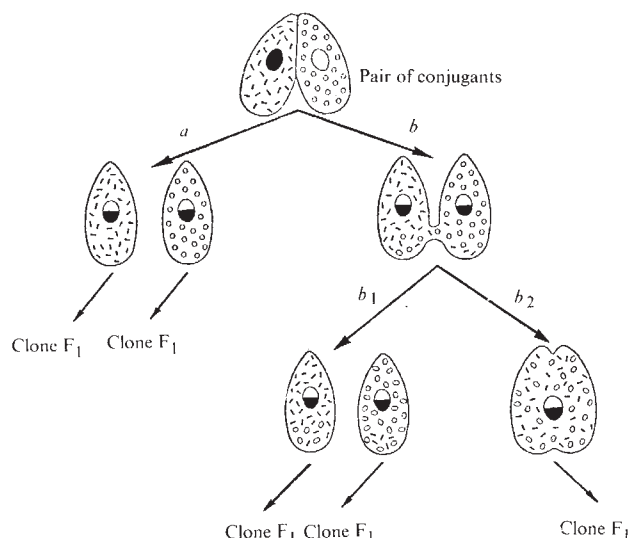


Fig. 1 Genetic exchanges between conjugating paramecia. The different fates of a pair of conjugants are represented. *a*, After completion of nuclear exchanges the two conjugants usually separate without cytoplasmic exchange. *b*, In some cases, a cytoplasmic bridge develops by the end of the conjugation process, through which cytoplasmic material is exchanged. *b*₁, After a variable time (min to h), the bridge is broken, the two conjugants separate and the relative amount of exchanged cytoplasm depends on the duration and width of the bridge. *b*₂, In rarer cases the bridge persists and eventually yields complete fusion of the two mates that reproduce true to type as a doublet.

Whenever an antibiotic sensitive cell that has just undergone a short cytoplasmic exchange with a resistant partner is placed in the corresponding selective medium, it first has a sensitive phenotype. Then, after a lag, it becomes progressively resistant and reaches full resistance after about four fissions. The lag is inversely proportional to the duration and size of the bridge. This observation, first made for $E^R \times E^S$ crosses¹¹, has been extended to all combinations studied (Table 1), amounting altogether to 200 independent heteromitochondriotes from fifteen different crosses. Conversely, a thermosensitive antibiotic-resistant ex-conjugant, when subjected to 36° C, turns progressively thermoresistant and becomes antibiotic-sensitive.

This phenotypic evolution is associated with a rapid genetic

purification for the selected mitochondrial character. Whenever an originally cytoplasmically mixed cell has reached ten fissions in a given selective condition, it becomes irreversibly genetically pure for the selected character. Systematic tests have shown that, after three fissions in erythromycin, cells which originally contained a minority of erythromycin-resistant mitochondria had often become "purely" erythromycin-resistant. From these results we infer that, during the process of acquisition of resistance under a given selective pressure, there is a differential multiplication of resistant mitochondrial genomes along with an elimination of functional sensitive genomes.

Table 1 List of Analysed Crosses

Cross	Analysis on Ex-conjugant from pairs with cytoplasmic bridge	Doublet cells
$E_1^R \times E_{(401)}^S$	10	1
$E_3^R \times E_{(401)}^S$	2	
$E_{102}^R \times E_{(401)}^S$	5	3
$E_{37}^R \times E_{(401)}^S$	6	
$E_{38}^R \times E_{(401)}^S$	6	
$C_2^R \times E_{(111)}^S$	3	9
$C_4^R \times E_{(401)}^S$	3	
$E_1^R T_1^R \times E_{(401)}^S$	10	
$E_1^R T_1^R \times E_{(401)}^S$	4	
$C_3^R \times E_{(401)}^S$	4	
$E_{37}^R \times E_{(401)}^S$	4	
$C_2^R \times E_{(401)}^S$	3	
$C_4^R \times E_{(401)}^S$	3	3

The figures indicate the number of ex-conjugant clones (or doublet cells) extensively studied and tested, at least at the first, tenth, twentieth and thirty-fifth post-conjugal fission.

All the strains belong to syngen 4 of *P. aurelia*. $E_{(401)}^S$ and $E_{(111)}^S$ derive from stock d₄₋₂ (Dr Sonneborn's collection), and are marked by thermosensitive recessive gene 401 or 111, used to ascertain that normal nuclear exchanges have occurred at conjugation. E_1^R , E_3^R , E_{37}^R , E_{38}^R were induced by ultraviolet irradiation, and are cytoplasmically inherited, erythromycin resistant and thermosensitive, the thermosensitivity being associated to erythromycin resistance¹¹. E_{102}^R is a spontaneous mutant, highly erythromycin resistant and only slightly thermosensitive. C_2^R and C_4^R are respectively an ultraviolet induced and a spontaneous chloramphenicol resistant mutant; both are thermoresistant. C_3^R , supplied by Dr G. H. Beale, is a chloramphenicol resistant mutant isolated in stock 51, after nitroguanidine mutagenesis; it is slightly thermosensitive. $C_2^R E^R$ is a doubly resistant strain, isolated from the C_2^R strain, and highly thermosensitive. $E_1^R T_1^R$ is a partial revertant from E_1^R , obtained after ultraviolet mutagenesis, that retains erythromycin resistance and has lost thermosensitivity.

Various pairwise combinations between resistant and sensitive mitochondria or between two types of resistant mitochondria have been followed under supposedly non-selective conditions (26° C, absence of antibiotic). Two types of results were obtained, some of which are recorded in Table 2: either the two types of mitochondria show identical "stability" in the mixed population and the cells remain "mixed" for a great number of generations (more than eighty for $C^R \times C^S$ combinations), or one of the two types of mitochondria, always the same for a given combination, is progressively eliminated and "pure" cells of the favoured mitochondrial type are recovered between the twentieth and the fortieth generations (all $E^R \times E^S$ and $C^R E^R \times C^S E^S$ combinations). In the second case, the wild type mitochondrial type is always the more stable. Moreover, among the various mutants studied, a classification can be made according to their relative degree of stability in heteromitochondriotes: $C_3^R > E_{102}^R > E_1^R > E_{37}^R - E_{38}^R > C_2^R E^R$. This classification parallels the classification of the mutants by

order of increasing thermosensitivity, C^R mutants being thermoresistant and $C^R E^R$ mutants being highly thermosensitive. Thus it is always the more thermosensitive allele which is eliminated in a mixed population. In good agreement with this classification, in $C^R \times E^R$ heteromitochondriotes, the C^R mitochondria take over the E^R ones just as the sensitive ones take over the E^R ones.

From the above results and some detailed pedigree analysis, we were led to two conclusions. (1) The number of resistant and sensitive "units" in each cell is high, as would be expected from the large number of mitochondria present in each cell (several thousands). Statistical analysis giving estimates of the number of mitochondria and details of pedigrees will be given elsewhere. (2) Each mutant displays a specific rate of transmission in mixed mitochondrial populations. This is an intrinsic property

Finally, several combinations between resistant markers have been extensively studied in order to detect recombination and/or complementation between mitochondrial genomes (two $C^R \times E^R$, two $E^R \times E^R$, one $C^R E^R \times C^S E^S$ and one $E^R TR \times E^R TS$ combinations). In no case did the simultaneous presence of two types of mitochondria result in enhanced resistance to a double-selective condition, although it was ascertained that the cells were indeed mixed. No complementation could thus be detected. Neither doubly sensitive nor doubly resistant recombinants were detected in the $C^R \times E^R$ combinations, despite the supposedly efficient screening procedure that was used and no recombinants were detected in any of the other combinations.

On the whole, mitochondria in *Paramecium* appear as autonomous, with respect to each other, for their replication and

Table 2 Evolution of Various Mixed Mitochondrial Populations in Non-selective Medium

Origin of the population	Type of combination	Fission number									
		10	20	30	40	50	60	70	80		
Pairs with a bridge	$E_1^R \times E^S$	$\begin{matrix} e \times S \\ e \times R \end{matrix}$	$\begin{matrix} 6/12 \\ 15/15 \end{matrix}$		$\begin{matrix} 2/15 \\ 15/15 \end{matrix}$		$\begin{matrix} 15/15 \\ 9/15 \end{matrix}$	$\begin{matrix} 0/30 \\ 0/30 \end{matrix}$			
	$E_{102}^R \times E^S$	$\begin{matrix} e \times S \\ e \times R \end{matrix}$	$\begin{matrix} R \\ R \end{matrix}$	$\begin{matrix} R \\ R \end{matrix}$	$\begin{matrix} 6/25 \\ 25/25 \end{matrix}$						
	$E_{38}^R \times E^S$	$\begin{matrix} e \times S \\ e \times R \end{matrix}$	$\begin{matrix} 3/3 \\ 15/15 \end{matrix}$		$\begin{matrix} 2/30 \\ 30/30 \end{matrix}$	$\begin{matrix} 0/30 \\ 0/30 \end{matrix}$					
	$C_2^R \times C^S$	$\begin{matrix} e \times S \\ e \times R \end{matrix}$	$\begin{matrix} R \\ R \end{matrix}$	$\begin{matrix} R \\ R \end{matrix}$	$\begin{matrix} 14/15 \\ 15/15 \end{matrix}$	$\begin{matrix} 9/15 \\ 15/15 \end{matrix}$	$\begin{matrix} 16/27 \\ 26/26 \end{matrix}$	$\begin{matrix} 27/27 \\ 27/27 \end{matrix}$	$\begin{matrix} 1/9 \\ 9/9 \end{matrix}$	$\begin{matrix} 0/9 \\ 6/6 \end{matrix}$	$\begin{matrix} 0/9 \\ 9/9 \end{matrix}$
	$C_2^R E^R \times C^S E^S$	$\begin{matrix} e \times S \\ e \times R \end{matrix}$	$\begin{matrix} 0/30 \\ 30/30 \end{matrix}$		$\begin{matrix} 0/30 \\ 0/30 \end{matrix}$						
Doublet cells	$E_1^R - E^S$		$\begin{matrix} 587/587 \\ 100\% \end{matrix}$	$\begin{matrix} 32/259 \\ 11\% \end{matrix}$							
	$E_{102}^R - E^S$		30/30	39/44	8/42	0/30					
			30/30	36/40	14/40	0/30					
			30/30	38/115	15/40	0/30					
			100%	90%	30%	0%					
	$C_2^R - C^S$		14/15	13/14	15/15		2/15		2/9		
			15/15	14/14	14/15		3/15		2/9		
			13/15	14/14	15/15		9/15		5/9		
			15/15	12/14	15/15		9/15		9/9		
			15/15	12/14	15/15		4/15				
			15/15	13/13	15/15		2/15				
			100%	100%	100%		32%		R		

The figures indicate the number of resistant cells over the number of tested cells, in clones issued from mixed cells, at various stages of growth in non-selective medium. For pairs that have undergone a bridge, numbers are given only for the 2 ex-conjugant clones of one representative pair. "R" indicates that only one to three cells have been tested and were found resistant. It is worth pointing out the close similarity of evolution of different doublet clones from the same cross in spite of the drift introduced by the serial subcloning.

of the given mutation, for all the strains used are isogenic for their nuclear genome, and therefore all the differences in behaviour recorded can be attributed to differences in mitochondrial genotype. This specific rate of transmission is analogous to what has been defined in yeast as the "polarity of transmission" of mitochondrial markers^{8,9}. In yeast, this polarity reflects, at least partly, recombinational events between mitochondrial genomes. In *Paramecium*, no such event has been detected. The good correlation between the rate of transmission of a resistant mitochondrial type and the "health" of the corresponding resistant cells, as judged by their growth rate and thermosensitivity, would rather suggest that the observed evolution of a mixed population reflects the selective advantage of one of the two types of mitochondria. This implies that, within a cell, the two different types of mitochondria "replicate" at a different rate.

for the expression of the characters they harbour, at least for the particular mutations studied, and in the precise genetic background used.

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Mitochondrial Genetics in *Paramecium*

RESISTANCE to erythromycin and chloramphenicol in yeast has been shown to be controlled by non-Mendelian genetic factors in mitochondrial DNA^{1,2}. Because *paramecium* has particular advantages for the study of cytoplasmic genetics³, we have made some genetic and biochemical studies on drug-resistant variants of this ciliate, working with syngens 1 and 4 of *P. aurelia*.

When *paramecia* are placed in bacterized lettuce medium (pH 6.8–7.0) containing 0.25 mg ml.⁻¹ erythromycin, growth stops after one to three fissions, but thereafter the ciliates remain alive for a considerable time. Between two and four weeks afterwards, erythromycin-resistant mutants appear^{4,5}, at a frequency of about 1 per 1,000 *paramecia* initially present. *Paramecia* resistant to chloramphenicol (0.25 mg ml.⁻¹) have also been obtained, but only after treatment with a mutagenic chemical (nitrosoguanidine). The variants have been shown to be stable for between one and two years when grown in media lacking antibiotics.

Genetic tests have shown, both in syngens 1 and 4, that resistance to both drugs is inherited through the cytoplasm at conjugation. When there is no cytoplasmic mixing at conjugation between sensitive and resistant *paramecia* (the normal situation), one of the ex-conjugant clones consists of resistant animals and the other sensitive. When there is cytoplasmic mixing, which occurs commonly in stock 51 (syngen 4), but rarely in other stocks unless induced by special treatments⁶, both ex-conjugants produce drug-resistant animals. The cytoplasmic basis of inheritance of erythromycin resistance was confirmed by a programme of repeated back-crossing to sensitive animals. No effect of nuclear genes on drug resistance has been shown, but the matter has not been intensively studied.

Cytoplasmic mixing between erythromycin-resistant and chloramphenicol-resistant conjugants results in the production of *paramecia* capable of growing in either antibiotic, but not in medium containing both. Such "mixed" clones, after growth in one antibiotic for a few (ten to fifteen) fissions, lose their resistance to the other antibiotic. When grown in drug-free medium for a hundred or more fissions, however, "mixed" clones may retain their capacity to grow in the presence of either antibiotic, although lines containing animals resistant to erythromycin alone appear sporadically. Thus in one experiment, six hybrid clones containing mixtures of cytoplasm from stocks 51-E^R (EDIN) (erythromycin-resistant) and 51-C^R (EDIN) (chloramphenicol-resistant) were grown in normal medium. Chloramphenicol resistance was lost after forty-seven fissions in one clone, after 106 fissions in three clones and after 195 fissions in the sixth clone. Erythromycin resistance was not lost at all in this experiment.

Similar experiments were done with clones containing mixtures of resistant and sensitive cytoplasm and the results

indicated that there was a slight selective advantage of "wild type" or sensitive (51-E^SC^S) cytoplasm over 51-E^R and of 51-E^R over 51-C^R, but these are probably strain or mutant-specific characteristics, judging by the different results obtained by Adoutte and Beisson⁷.

Attempts to obtain "recombination" between 51-E^R (EDIN) and 51-C^R (EDIN) cytoplasmic components by selection of doubly resistant animals have so far failed, although such doubly resistant types have been obtained by mutation.

In an attempt to identify the particular cytoplasmic factor associated with the transfer of antibiotic resistance, various cell fractions were prepared from erythromycin-resistant *paramecia* (168-E^R (EDIN)) and injected into sensitive *paramecia* (168-E^S (EDIN)) using a micro-injection technique⁸.

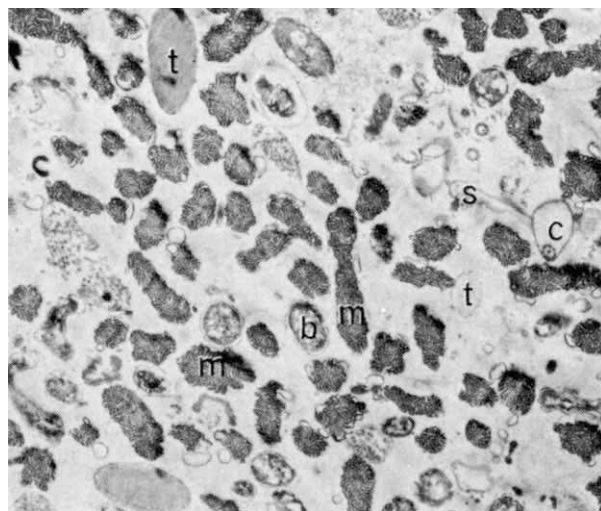


Fig. 1 Electron micrograph of an osmium fixed mitochondrial preparation, pelleted and embedded in 'Araldite'. M, Mitochondria; b, bacteria; s, fragments of surface membrane; c, cilia; t, trichocysts.

Two methods of preparing cell fractions for micro-injection into sensitive cells were used. (i) Packed cells were resuspended in four volumes of a sucrose buffer (0.3 M sucrose, 100 mg ml.⁻¹ bovine serum albumin, 0.001 M potassium phosphate, pH = 7.0) at 4° C and were twice homogenized in a cream homogenizer. After centrifugation at 1,500g for 7 min, the supernatant, which would be expected to contain most cell components except macronuclei and large pieces of pellicle, was used for micro-injection. Of a total of thirty-four surviving cells injected with this type of preparation, twenty-three developed the ability to grow in erythromycin after a delay of 9–15 days at 24° C. None of the seventy-four uninjected controls became resistant. (ii) Mitochondria were prepared by differential centrifugation in raffinose buffer⁹. The centre section of the "mitochondrial" pellet was resuspended in the smallest possible volume (about 0.5 ml.) of the raffinose homogenization buffer and injected into *paramecia* as soon as possible.

An estimate of the composition of this preparation was obtained by examination of an osmium-fixed pellet under the electron microscope (Fig. 1). The following percentages of various particulate components were found: mitochondria, 80%; bacteria, 10%; fragments of surface membrane, 4%; cilia, 1%; trichocysts, 4%; other, 1%. These percentages represent the numbers of each component in relation to the total number of particles counted.

Of sixty-four animals injected with this preparation, fifty-four became resistant after 4–15 days while none of the 132 uninjected controls developed resistance. Five cells injected with the post-mitochondrial supernatant and eleven cells injected with

preparations from sensitive cells did not become resistant. The spontaneous mutation rate is of the order one in 10^3 cells (one in 10^7 mitochondria), so these results are felt to be highly significant. It is also important to point out that positive results were only obtained following the use of an injection needle with an internal diameter of not less than $3.0\ \mu\text{m}$ and that this corresponds well with the size of the mitochondria of paramecium ($2\text{--}4\ \mu\text{m}$ diameter). Although the injected material did not consist exclusively of mitochondria, our results are entirely consistent with the view that transfer of resistance from one cell to another was in fact mediated by these organelles.

Our further work has concerned the site and biochemical nature of the alterations produced in drug-resistant paramecia. Erythromycin and chloramphenicol are both known to inhibit protein synthesis on bacterial ribosomes, and alterations in the ribosomal proteins have been shown to occur in erythromycin-resistant strains¹⁰, so we have investigated the possibility of similar alterations in the mitochondrial ribosomal proteins of drug-resistant paramecia.

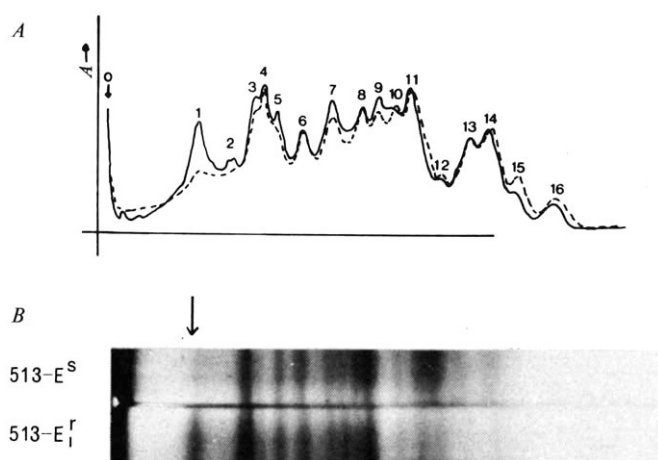


Fig. 2 *a*, Comparison of densitometer traces of stained acrylamide gels of separated "crude" mitochondrial ribosomal proteins from strains 513-E^S (EDIN) (---) and 513-E^R (EDIN) (—). The proteins are numbered 1–16 from the origin, marked ○. *b*, Photograph of the acrylamide gels scanned in *a*. The difference between the two strains in band 1 is indicated by the arrow.

Twice-washed mitochondria were prepared by differential centrifugation⁹ and then lysed by homogenization in 2% 'Triton X-100 TMK' (50 mM Tris-HCl, pH 7.5, 10 M MgCl₂, 10 mM KCl), using a 'Teflon' glass homogenizer. The mitochondrial ribosomes were separated by differential centrifugation of this lysate¹¹ and the proteins extracted from the crude ribosomal pellet by treatment with 4 M LiCl–8 M urea at 4° C for 12 h, the RNA being removed by centrifugation after this treatment¹². The ribosomal proteins from resistant and sensitive strains were then compared by acrylamide electrophoresis at pH 4.5^{12,13}. Electrophoresis was carried out for 4 h at 4 mA gel⁻¹ at 4° C. The gels were stained with 1% naphthalene black for 1 h, destained electrolytically in 7% acetic acid and scanned using a Joyce-Loebl ultraviolet scanner.

The electrophoretic pattern of these proteins shows some sixteen bands (Fig. 2) and differences have been observed between sensitive and drug resistant strains. For example, comparison of 513-E^S (EDIN) (syngen 1, sensitive) with 513-E^R (EDIN) (syngen 1, spontaneously erythromycin-resistant) shows a quantitative alteration in a single protein band (No. 1) (Fig. 2). The difference observed was found consistently in different electrophoretic runs and using independent preparations of the crude ribosomal proteins from the two strains. The ratio of the area under peaks 3 and 4 to the area

under peak 1 has been calculated for the two strains and the difference between this ratio for 513-E^S (EDIN) and 513-E^R (EDIN) was shown to be significant. While it is realized that the preparation of mitochondrial ribosomal proteins is crude, these results are consistent with the view that the change from sensitivity to erythromycin resistance involves the alteration of one or more proteins associated with the mitochondrial ribosome.

The precise nature of this difference has not been ascertained but various possible explanations are now being investigated. Attempts are also being made to establish unequivocally the mitochondrial ribosomal origin of this protein.

Our results show that erythromycin resistance and chloramphenicol resistance in *P. aurelia* are due to cytoplasmic genetic factors located in the mitochondria (confirming the findings with yeast). These mitochondrial "genes" modify certain proteins associated with the mitochondrial ribosomes. The special features of the paramecium material, such as the very large size of the "cells", permitting transfer of mitochondria by micro-injection, the fact that the reaction of individual cells to the drugs can be studied, and the large size and number of mitochondria per cell (as compared with yeast) will make possible the elucidation of new features of mitochondrial genetics. Further technical and other details of this work will be published elsewhere.

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Stimulus Characteristics of Marijuana Components

THE effects of certain drugs are sufficiently strong and distinctive for animals to learn differential responses solely on the basis of whether they are in the drug or control condition¹. Tests with lower doses of the same drug can establish the threshold for this discriminative response, and tests with other compounds can be used to determine the similarity or dissimilarity of their subjective effects. We have used this method for characterizing the pharmacological actions of Δ^1 -tetrahydrocannabinol (Δ^1 -THC), which is reported to be the major psychoactive constituent of marijuana²⁻³.

Twelve male albino Wistar rats, housed in individual cages, were maintained on a food-deprivation schedule of 12 g of

Purina Lab Chow checkers daily, with water constantly available. The Δ^1 -THC (or Δ^9 -THC by an alternative nomenclature) was prepared and stored as described elsewhere⁴⁻⁵. The animals were trained to associate the drug and control conditions with opposing approach and avoidance responses⁶. A lever-pressing approach response was established by delivery of a food pellet (45 mg) after every fifth lever press throughout the 5 min sessions. Each session began 30 min after injection of Δ^1 -THC (4 mg/kg, intraperitoneally) for half the animals and the control fluid for the others. In approximately half the sessions, the treatment was reversed (control instead of Δ^1 -THC or Δ^1 -THC instead of control), and every fifth lever press was followed by punishment (painful electric shock) instead of food reward. Approach or avoidance response was defined by whether the animal completed at least five responses during the 5 min session. The sessions of reward and punishment, associated with the drug and control conditions, were given in a varied sequence with an interval of one or more days between successive sessions.

One animal, trained to avoid in the control condition, consistently avoided also after drug injection and was dropped from the study. The remaining eleven rats averaged more than 90% correct responses (approach in the rewarded condition and avoidance in the punished condition) throughout 105 sessions after preliminary training. Interspersed among these 105 sessions were fifty tests in novel drug conditions, according to procedures described previously⁷. Choice of the Δ^1 -THC response was signified by approach for half the animals and avoidance for the others.

each preparation by gas-liquid chromatography. Each of the four preparations showed a consistent dose-response function. Drug responses close to 100% or 0% necessarily mean approach by some animals and avoidance by others. The intermediate percentages also were fairly evenly divided between these two categories, rather than due to avoidance or approach by all animals regardless of which type of response was associated with Δ^1 -THC.

These findings indicate that important subjective characteristics of marihuana can be reproduced by Δ^1 -THC, which supports the claim that Δ^1 -THC is the principal psychoactive component of marihuana²⁻³. A synergistic action of Δ^1 -THC with certain other compounds, such as the *n*-propyl analogue of Δ^1 -THC recently identified as a component of marihuana extract⁸, is suggested by reliably greater potency of the alcoholic marihuana extract than the other three preparations, which did not differ reliably from each other in this respect.

A generalized metabolic inhibitor, SKF 525-A (25 mg/kg, intraperitoneally), elicited 0% drug response by itself but 100% drug response when followed by Δ^1 -THC (1 or 4 mg/kg). This enhancement of the drug response adds to other evidence⁹ against a suggestion¹⁰ that the active compound may be a metabolite rather than the Δ^1 -THC itself.

Tests were conducted with various other pharmacological agents, including major and minor tranquilizers (chlorpromazine and chlordiazepoxide), central nervous system depressants (pentobarbital and ethyl alcohol), psychotomimetics (mescaline, dimethyltryptamine, and LSD-25), anticholinergics (atropine and scopolamine), other cannabinoids (cannabinol and cannabidiol), morphine, and cocaine. In all cases, there was evidence for a dissimilar subjective effect from that of Δ^1 -THC. This highly specific response to Δ^1 -THC may be a useful device for distinguishing it from other types of drug, because the observable effects of Δ^1 -THC and marihuana, such as central nervous system depression and hyperexcitability¹⁰, are elicited also by various physiological and environmental conditions.

This research was supported by a Public Health Service training grant from the NIGMS (for R. K. K.) and a research grant and research scientist development award from the NIMH (to H. B.). The Δ^1 -THC synthesized by ADL and marihuana extract distillate were received from the NIMH-FDA Psychotomimetic Drugs Committee. Technical assistance in analysis of compounds by gas-liquid chromatography was received from Dr Micha Theiner and Dr Alvin B. Segelman.

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Table 1 Responses of Rats to Treatment with Δ^1 -THC compared with Other Drugs

Δ^1 -THC mg/kg	Δ^1 -THC synthesized by Mechoulam (97% pure)	ADL (94% pure)	Marihuana extract distillate (17% Δ^1 -THC)	Alcoholic marihuana extract (0.4% Δ^1 -THC)
16	100	—	—	—
4	93 ‡	91	82 *	—
2	77 *	73	60	—
1	33 ‡	0	10	91
0.50	0	—	—	45
0.25	—	—	—	9
0	8 ‡	—	—	—
ED ₅₀ (mg/kg)	1.40	1.88	1.94	0.51
95% confidence limits	0.93-2.12	1.26-2.79	1.02-3.53	0.32-0.80

* Average of two tests.

‡ Average of three tests.

‡ Average of many training sessions.

Percentage Δ^1 -THC response (approach for half the animals, avoidance for the others) in tests with several doses and preparations containing this compound by rats trained to make differential responses to 4 mg/kg (synthesized by Mechoulam) and the non-drug control condition (0 mg/kg). All data are for eleven animals except that ten were tested with marihuana extract distillate at the two lower doses and in one test at the highest dose.

Table 1 shows the percentage of drug response in tests with Δ^1 -THC or preparations of marihuana containing this compound. All training sessions were conducted with Δ^1 -THC provided by R. Mechoulam. An adaptation of standard probits⁷ was used for calculating ED₅₀ and 95% confidence limits. In addition to tests with a wide range of doses for the Δ^1 -THC used for training, tests were conducted with the same compound synthesized by Arthur D. Little, Inc. (ADL), with marihuana extract distillate, and with an alcoholic marihuana extract. We determined or verified the Δ^1 -THC content in

BOOK REVIEWS

Kafka Country

A Question of Madness. By Zhores A. Medvedev and Roy A. Medvedev. Translated from the Russian by Ellen de Kadt. Pp. 223. (Macmillan: London, November 1971.) £2.75.

IF a celebrated British scientist came to one saying that he believed that a colleague whose views he had refuted had conspired with the authorities to have him categorized as mentally disturbed and forcibly detained in a mental hospital, one's first reaction would almost certainly be to wonder whether the speaker was not indeed mentally disturbed. The most remarkable thing about this narrative of Zhores Medvedev's 19-day incarceration in a Russian district mental hospital is the growing conviction that such things *can* happen in that country. The book begins with a factual account of his being visited at home by the chief psychiatrist of the Kaluga District Hospital, who brought with him three policemen and forcibly hauled him off to a locked ward in his hospital. Both Zhores and his twin brother, Roy Medvedev, insisted vehemently that he was perfectly sane and that by no stretch of the imagination could he be regarded as having been a source of danger to himself or to others. During the days following his incarceration, Roy Medvedev rallied a large circle of friends, including numerous prominent Russian scientists and writers, who kept up a steady barrage of telegrams and representations both to the local hospital authorities and to officials in the Federal Ministry of Health.

The narrative reads like a thriller, each stage being carefully documented by notes which the brothers Medvedev kept at the time. It is quite a melodramatic story, in which the heroes and villains are painted in bold terms. From the beginning Dr Lifshits, the head doctor of the Kaluga Psychiatric Hospital, comes out in a very unflattering light. The authors believe him to have been the subservient agent of authorities in the Ministry and the Party. He certainly did not stand up well to insistent questioning about the grounds on which he had certified Zhores as being in need of compulsory treatment, and he repeatedly gave assurances of his impending release, which he later failed to implement.

Early in the course of events, Roy Medvedev sought the intervention of the USSR Ministry of Health and gives a very unflattering report of the reactions of Dr Serebryakova and Professor Snezhnevsky, two of the top psychia-

trists in that Ministry. In a thumb-nail sketch of the latter's rise to power, after the anti-semitic campaign which took place during Stalin's last years, Roy Medvedev says: "Psychiatry was now monopolized by the school of A. V. Snezhnevsky, who announced a new unitary 'Pavlovian theory of schizophrenia'. Snezhnevsky succeeded in foisting his much too broad classification of mental illness on Soviet psychiatrists, but this was repudiated by the World Health Organization". This author is equally uninhibited in denouncing a number of Soviet psychiatrists, including Dr Lunts "of the notorious Serbsky Prison Hospital", Dr G. Morozov and Dr A. A. Portnov. Dr Portnov is described as "professionally untrustworthy and morally pliable".

There are heroes as well as villains. For example, Roy Medvedev describes a psychiatrist named L., recommended to him by his friends as "an experienced and honest doctor", and states "as soon as I met L., I felt I could trust him completely".

There are also many instances of disinterested intervention on the part of other Soviet intellectuals, including A. D. Sakharov, Tvardovsky and A. Solzhenitsyn; but not every such intellectual responded so generously. Dr V. I. Khozinsky responded to an American colleague by quoting—which he had no right to do—details of the medical diagnosis used by Dr Lifshits, and implying his acceptance of this diagnosis. Zhores Medvedev comments on this letter: "Khozinsky may well have believed what he wrote in his letter but there can be no doubt whatsoever of his utter dishonesty and lack of decency".

This polarization of the protagonists into the wholly good and the utterly bad is not necessarily idiosyncratic to the brothers Medvedev; it was described as a very general trait in the study of Russian national character published by Geoffrey Gorer and John Rickman in 1949.

It is a mark of all advanced civilizations that the rights of their individual citizens are recognized, and protected by legal safeguards. The vital importance of such safeguards is brought home to us by the injustices perpetrated daily by police states such as South Africa. The USSR recognizes the need to protect individual liberty in principle; but this case forcefully raises the question whether in practice these liberties have been eroded in recent years by increasing resort to psychiatric "diagnoses" and compulsory "treatment" as a means of

discrediting embarrassing political critics.

There is a long tradition in Russia for such abuses of psychiatry. Under Tsar Alexander I a military cadet named Zhuko was declared insane because he wrote a collection of verse about freedom. Under Tsar Nicholas I the philosopher Chaadaev, a friend of Pushkin, was decreed insane and confined to his house for a year after publishing an article criticizing tyrannical actions of the Tsar's regime.

In recent years, however, there have been increasingly frequent instances in which persons—such as General Grigorenko and Ivan Yakimovich—who dared to criticize governmental actions have not been arraigned under Articles 70 ("anti-Soviet agitation and propaganda") or 190 (i) ("dissemination of deliberately slanderous fabrication defaming the Soviet state and society") because of the unwelcome publicity attending court trials, but have instead been categorized as mentally ill and subjected to compulsory treatment.

These authors write with strong feeling, which finds expression in a number of hasty and sweeping judgments. They have two main aims: to demonstrate that Zhores Medvedev was improperly and unjustly incarcerated, and to warn against the wider dangers of the abuse of powers of psychiatric detention. There are many involuntary indications that Zhores may have been—and may still be—an extremely awkward, impulsive, stubborn and litigious individual. But all this is irrelevant to the key issue, namely, whether his behaviour constituted a danger to himself or to others; and although Zhores's conduct may have been exasperating to the numerous colleagues and local Party officials with whom he disagreed, there is nothing to suggest that his conduct justified his being treated compulsorily.

Protection against the misuse of compulsory treatment depends on three factors: (i) the formulation of regulations designed to protect the interests of the patient, (ii) the personal integrity of the psychiatrists involved, and (iii) the existence of a public opinion which can be mobilized in defence of patients' civil liberties. In this instance, Roy Medvedev had great difficulty in gaining access to the regulations governing compulsory treatment: he refers to them with characteristic impetuosity as "the notorious regulations" but he also quotes them in full and reveals them to be reasonable. Their opening clause states: "If a mentally ill person is clearly a danger to himself or those around him the health authority have the right to

place him in a psychiatric hospital without the consent of the ill person himself or his relatives or guardians". Roy Medvedev himself repeatedly reproaches the Kaluga Hospital psychiatrists because they have not behaved strictly in accordance with these regulations; but he rashly suggests that the latter should specify in precise terms "all possible grounds known to psychiatry which indicate compulsory hospitalization".

This is precisely where the Kaluga psychiatrists were at fault. They tried to justify their actions by "diagnosing" Zhores as suffering from "paranoid tendencies" or "poor adaptation to the social environment"; but the decision to institute compulsory treatment should be determined, not by the psychiatric diagnosis but by the patient's actual behaviour, which can be described in simple, non-technical language.

The brothers Medvedev do not hesitate to impugn the integrity of the Kaluga psychiatrists who had agreed to "take on the chief parts in the scenario"; indeed they denounce every psychiatrist who appears willing even to consider the possibility that Zhores may be somewhat unbalanced. At best, they are depicted as the venal or cowardly agents of Zhores's unseen enemies, who are believed to have engineered the whole affair from behind the scenes. Roy Medvedev writes: "I never did find out, however, exactly which institution had sought their help in order to put an end to Zhores's criticism. Perhaps it was the Lysenko institutions which still exist in our country? Or the Censorship Department of the Post Office? Or the organization responsible for international scientific exchanges and tourism".

Many visitors to the USSR will recognize, from the experience of their scientific colleagues, this mood of impotent speculation as to just who it is within the "apparatus" that makes the vital decisions concerning censorship, or the right to publication, or permission to travel abroad.

Perhaps the most striking feature of this book, however, is the clarity with which it reveals the existence of a large body of opinion which is able and willing to challenge these faceless men who make arbitrary bureaucratic decisions. It becomes evident that the USSR is not simply a totalitarian state, but is to some extent already a pluralistic society. In the narrative, there is a constant tug-of-war between repressive and libertarian forces. For example, during an international symposium of geneticists which was being held in Moscow at this time, one of the Medvedevs' friends wrote on the blackboard in large letters: "ACADEMICIAN A. D. SAKHAROV IS IN THE AUDITORIUM COLLECTING SIGNATURES FOR A PROTEST

AGAINST THE COMMITMENT OF ZHOSES MEDVEDEV TO A MENTAL HOSPITAL". This at once attracted considerable support, but several young would-be signatories were advised not to sign, as this would get them into trouble; and after a little while two men in plain clothes came up to Sakharov and asked for his identity papers. He did not have them, so he had to leave.

Evidently, there are many intellectuals in Russia today who are prepared to speak out in defence of their fellow-citizens' freedom of conscience: Roy Medvedev specifically cites many Soviet psychiatrists who vigorously repudiate the abuse of their medical role for political ends; and he cites the particularly eloquent open letter, entitled "This is how we live", in which Solzhenitsyn denounced Zhores's incarceration and all similar attempts to stifle honest expressions of opinion. In his concluding chapter, Roy Medvedev widens the debate. It is not merely, he says, this particular form of repression which has to be opposed, but every attempt to suppress and stifle opinions distasteful to the country's rulers. He asserts: "A society cannot be genuinely democratic if the right of the majority to govern is not secure, if the majority is unable to take decisions and implement them. However, neither can there be genuine democracy if the right to express and defend minority opinion is also not protected. . . . The right of dissent should not be thought peculiar to bourgeois democracy. It is a most important feature of any democracy".

It is surely heartening to learn that liberal opinions can redress instances of personal injustice in the USSR today, and to read such a forthright defence of independence of thought; but it is chastening to be reminded that the Medvedevs' book had to be smuggled out of their country and published abroad. The battle for intellectual freedom has not been won yet in the USSR—nor can its defence be relaxed in any of the countries of the "free" world, for here, too, there is a constant struggle between moral and intellectual conservatism and that "divine discontent" which subjects every accepted belief to critical evaluation. The Medvedevs' story is a parable for all mankind.

G. M. CARSTAIRS

Industrial Relics

The Chemical Industry. By W. A. Campbell. Pp. 156+25 plates. (Longman: London, September 1971.) £3.

ABOUT a decade ago it became apparent that a process initiated by Hitler's air force and continued by well-meaning planners and developers might leave Britain with few relics of the days when she truly was the workshop of the

world. So was born industrial archaeology. It initiated a flood of publications dealing with the more obvious remains of our industrial evolution. Many of these were indeed "industrial monuments"—great works of civil engineering associated with canals, railways, mills, and so on. Although many are gone their intrinsic aesthetic merit meant that they survive in engravings and paintings because artists like Turner and Loutherboung saw in the industrial scenes of their day a new romanticism which they recorded, often apparently emphasizing the pollution the new processes were causing. The scholarship displayed in industrial archaeology publications has varied greatly in quality, understandably, because the knowledge required to place an industrial relic in its total social, technical, and economic setting straddles a wide range of disciplines. This may explain in part the apparent neglect of the chemical industry as a hunting ground for the new archaeologists. But discreetly tucked away towards the end of W. A. Campbell's book there is another explanation.

"It must be conceded that the chemical industry does not readily furnish material for the industrial archaeologist. Obsolete chemical plant has never inspired the same sentiments as veteran motor cars or locomotives, and, where detailed accounts survive, the end of a once-prosperous works is too often summed up in the final phrase 'sold for scrap'. The waste heaps, too, have not been matters for local pride: the communities which engaged in costly litigation over loss of amenity were not likely to spend more money on the conservation of monuments of pollution. However, the recent trend towards the recovery and landscaping of derelict industrial land provides an opportunity which the student of the chemical industry should not neglect."

It is in this light that we must read any book on the industrial archaeology of the chemical industry: written records are often easier to find than physical remains, so archaeology proper yields place to industrial history. Gone are the sites where sulphuric acid, that key chemical, was first made in industrial quantities; gone too is Tennant's 450-foot stack built to dispel the hydrogen chloride from his LeBlanc furnaces at St Rollox, Glasgow, in the days before the Alkali Acts. Transformed is much of the Swansea Valley, once the scene of utter desolation. Indeed it is worthy of record that so great is the change in our attitude to pollution that, while the British Association advertised its 1880 meeting in Swansea with a poster that may aptly be described in one Langford's words as "the landscape of Hell foreshadowed", on an excursion arranged for the 1971 meeting it was late in the

afternoon before the excursionists found anything archaeological to examine.

This is not to imply that the author's book is not a valuable addition to the literature of a rapidly growing subject. Even viewed as a short history of the chemical industry it is full of clues to follow up. Of particular value to the historian of technology are the later parts of the book with its hints on "where to find the facts", a table of works registered under the Alkali Act of 1863 (and we must remember that parliament's idea of an alkali is quite different from that of a chemist), a gazetteer locating the germinal foci of ICI's vast empire, and giving the locations of about a score of existing chemical firms founded more than a century ago.

A modern poet might wax lyrical over a cat-cracker; no early chemical works was a thing of beauty, but if W. A. Campbell's book does nothing more than stimulate interest to get more information about the origins of our industrial history on to the files, and existing archives into safe keeping, it will serve a very useful purpose.

ARCHIE CLOW

Behavioural Biology

The Biological Bases of Behaviour. By Neil Chalmers, Roberta Crawley and Steven P. R. Rose, assisted by Judy Hicklin. Pp. 318. (Harper and Row: New York and London, November 1971.) (Published for the Open University Press.) £5 cloth; £2.75 paperback.

THIS book is a "reader"—a collection of reprints from the *Scientific American*, *Science Journal*, *Nature* and various other journals and books, chosen by the editors in relation to a second-level course organized by the Open University on the biological bases of behaviour. The articles cover a wide range of neurobiological subjects from the basic structure and function of the nervous system up to emotion, learning, intelligence and mind. The numerous authors include some of the best-known people in the field, for example, Eccles, Katz, Huxley, von Békésy, Hubel, Hess, Olds, Miller, Sperry, Lorenz, Sherrington, Adrian and others of comparable prominence. The standard of writing of the individual articles is generally high, as one would expect from their origins. The collection will provide excellent background reading for students unversed in biological matters and will also be of interest to many others who work in unrelated fields but who have an interest in brain and behaviour. The volume could perhaps have been improved by the addition to each article of a short list of up-to-date references.

To an extent, the editors have chosen

the easy way out in that they have compiled this book rather than written it. This method has the advantage that many of the papers are written by the leading people in the field (or, perhaps, rewritten by the staff of the *Scientific American*, after originals by the leading people in the field) and thus carry a certain authority. A disadvantage of the method is that, in the rapidly-moving world of neurobiology, several of the articles are bound to be out of date, in which case the authority lent by the author's name to what was originally an ephemeral work can be misleading.

Religion and politics both enter this book. And whereas it is arguable that, as manifestations of human behaviour, both subjects have a good place in such a work, they seem somewhat out of place in this particular context. Thus MacKay makes an intriguing attempt to demonstrate the existence of free will, using scholastic arguments involving paradox, while the political aspect is evident in some of the phraseology of the editors. Expressions such as "pseudo-scientific arguments" (p. 211), "To bolster political acts" (p. 211), "The segregationists of the southern United States" (p. 239), "The Powellite elements of the Tory Party" (p. 239) would be more at home in political tract than a text of biology.

The editors make a point of mentioning in the preface that they are concerned about the social implications of neurobiology, and this is all to the good. It would be a pity, however, to mislead students into thinking that it is scientific to state that all men are equal, while it is pseudo-scientific to state that some are more equal than others. Both statements are about on a par as far as their scientific nature is concerned; and the term "pseudo-scientific" in this context is normally to be translated as "contrary to what I believe".

R. M. GAZE

Mechanism and Vitalism

Interpretations of Life and Mind. Edited by Marjorie Grene. Pp. xvi+152. (Routledge and Kegan Paul: London, October 1971.) £2.25.

THIS report of the Study Group on the Unity of Knowledge contains eight papers presented to Michael Polanyi on his eightieth birthday. The sub-title is "Essays around the Problem of Reductionism", the aim being to combat dogmatic reductionism by developing a viable alternative, most of the participants being insufficiently aware that physics is already doing precisely that. The editor's contributions, a paper by I. Prigogine, and one or two others are useful, but some of them remind me of A. B. Johnson's warning (1836): "Much

of what is esteemed as profound philosophy is nothing but a disputatious criticism of the meaning of words". The main themes are reducibility; the meaning of "mechanism"; the relations of behaviour, belief, and emotion; a critique of artificial reason; and Polanyi's tacit knowledge. Time can be saved by using Dr Grene's introductory notes to guide one to the papers most likely to be of interest, and her up-to-date bibliography gives references on reducibility.

Marjorie Grene considers the proposition "Biology is reducible to physics" and takes it to mean that all biological laws are derivable from universal laws of matter in motion. But, as she is aware, the classical concepts of matter and mechanics have long been transcended, for example in Maxwell's electromagnetic field. In 1939 Eddington pointed out that "the concept of substance has disappeared from fundamental physics; what we ultimately come down to is *form*", that is waves, fields, symmetry, and structure. This is even more true today. Physical theory, though retaining the title quantum *mechanics*, has gone beyond mechanism and has become a mathematical doctrine of symmetry and its breakdown, levels of particles and forces, the statistics of observables, and obscure "interactions". Thus the "problem of reductionism" in its original form has vanished, for physics itself is evolving methods for describing structures "at new levels" with "new" non-classical properties.

This is illustrated by a paper that I can recommend to physical scientists: Prigogine on "The Unity of Physical Laws and Levels of Description". He reports recent work of his Brussels school indicating that systems sufficiently far from equilibrium may display fluctuations leading to the formation of "completely new space-time organization"—a new structural level, with corresponding "coherent behaviour". Those interested in such structures on the frontiers of thermodynamics should compare Prigogine's references with McClare's extension of the Second Law (*J. Theoret. Biol.*, **30**, 1; 1971). Though such very new ideas must be approached cautiously, it is noteworthy that both authors suggest that complex molecules, which may or may not be biomolecules, are distinguished in certain respects from their thermal environment; for example, by their much more rapid internal resonance processes. Thus it is becoming clear that the entropy principle, correctly understood, under certain circumstances permits the occurrence of coherent processes in stable units constituting a new level of structure, as indeed it must.

Dr Grene suggests that "reducibility" may be a side issue. It appears, how-

ever, that it is void of content unless reformulated with relevance to the changed scientific situation.

LANCELOT LAW WHYTE

Dislocation Theories

Fundamental Aspects of Dislocation Theory. (Proceedings of the National Bureau of Standards, April 1969.) Edited by John A. Simmons and R. de Wit. Vol. 1: Pp. xxv+726. Vol. 2: Pp. xi+729-1138. (US Government Printing Office: Washington DC, December 1970.) \$8.25 per set. (Sold in sets only.)

THESE volumes record a conference held in 1969 under the auspices of the Institute for Materials Research, the purpose of which was "to provide a multidisciplinary forum seeking to intermix the viewpoints of solid state physicists and continuum mechanicians on the basic properties of dislocations". The proceedings include seventy-five contributed papers and two panel discussions on diverse topics.

Reactions to the diversity of research described here must inevitably be subjective to some extent: I shall only mention certain contributions which appeal to me, and emphasize that others with a different outlook might validly select a completely different list from the many papers of high quality. My feeling is that significant advances in the understanding of structure-sensitive properties of solids have always arisen from an interest in particular phenomena, such as brittle fracture, work-hardening of metals, or crystal growth at low supersaturation ratios. Attempts to attain a high level of generality, following the example (and sometimes the methods) of relativity or elementary particle theory, have not been rewarded in the same way. Accordingly my criteria for evaluating the papers were these: first, does the subject refer to a general problem in the science of real materials, rather than mathematical development for its own sake? Second, does the paper contain significantly new results relatively insensitive to special assumptions (for example, the form of the interatomic potential)? Third, is the model realistic enough to make predictions in a form suitable for experimental verification?

The papers which strictly answered these criteria were some of those relating to Peierls stresses, activation phenomena, and interaction with electrons and phonons. H. Suzuki presented a particularly interesting paper on the marked effect of the atomic arrangement in the slip plane on the lattice force on dislocations, and the papers by Duesbery and Hirsch and by Foreman, Hirsch and Humphreys on the flow of crystals containing hard particles both compare detailed theoretical models

with fairly new experimental data. Those by Gruner and by Klemens on phonon scattering by dislocations also present specific results in an important field in which large effects due to dislocations have still not been satisfactorily explained. There were surprisingly few papers on the interaction of electrons and dislocations, and of these the contribution by Haasen and Schröter best seemed to me to measure up to the importance of the field: they show that a one-dimensional energy band theory of electron states along the dislocation line provides a better understanding of experimental observations on germanium and silicon than the "dangling-bond" picture.

I also, however, found stimulating a number of papers in the section on "Applications of the Geometry of Dislocations". S. Mendelson, for instance, gives a wide-ranging and useful discussion of possible effects due to non-planar dissociation of dislocations, including flow in hexagonal metals and in germanium. Most interesting of all were the papers on disclinations, those Volterra elastic singularities neglected until recently on account of their forbiddingly high energy in nearly perfect crystals. Nabarro provides the conference with a comprehensive review of the topological properties of screw disclinations, while Friedel and Kléman illustrate the application of disclination and dislocation theory to liquid crystals. W. F. Harris shows that the subject may be relevant to certain periodic structures in biological materials.

Altogether, then, I found much to admire in these proceedings, qualified by some disappointment. The reason for this was expressed by Professor Seeger in the second panel discussion, in which he rightly lamented that the contribution of the subject to technologically important fields of fatigue and fracture, or the understanding of yield criteria in macroscopic plasticity, has been, and seems likely to be, small. Since the hope of elucidating these complex matters provided the original stimulus of the subject (and much of the financial support since), these failures should serve as a challenge and a warning against complacency.

K. E. PUTTICK

Solid Surfaces

Tribology. By E. D. Hondros. Pp. 70. (Mills and Boon: London, October 1971.) £1.50.

THIS little book which occupies less than 70 pages is an attempt to provide the engineer with some of the basic ideas that lie behind the term tribology. One of the principal problems that the author has had to face concerns the standard of knowledge and the level of intelligence assumed in the reader. It cannot be said

that the author has been particularly successful or consistent in this difficult task. At some times the writing is stiff and technical, at other times oversimple. Again some of the figures seem of little interest to engineers—for example, the subsurface cracking in polished beryllium or germanium. On the other hand, one or two of the schematic diagrams communicate well.

In spite of the rather mixed quality of the book it makes clear the general point that solid surfaces have special topographical, structural, energetic and chemical properties and that these play a crucial part in the majority of sliding processes. Perhaps the most significant phrases of the whole book are to be found in the first couple of pages, where the author points out that the only new feature of tribology is its name and that unhappily the use of this new word will not automatically simplify the complicated phenomena involved in "interacting surfaces in relative motion and of related subjects and practices". Nevertheless a new emphasis has emerged: the recognition that tribology is not only a matter of science and technology. The solution of tribological problems also involves managerial and economic factors of considerable complexity.

DAVID TABOR

Mathematics for Models

Mathematics for Ecologists. By I. Chaston. Pp. 132. (Butterworth: London, November 1971.) £2.20 cloth; £1.50 paper.

THIS book aims to introduce biologists to the most elementary and most essential parts of calculus and matrix theory—easy ideas, but very useful in mathematical theories and models. There is also a fascinating chapter on operational research methods. The concepts are illustrated by interesting examples (even if artificial ones) which give a good insight into the power of mathematical methods. Unfortunately it is less easy to be complimentary about many of the explanations. For example, a determinant is first defined as follows: "If an operation is carried out whereby all the elements within a square matrix A are described by a single value . . . this calculated value is known as the determinant . . .". What is a novice to make of this? Later on this becomes ". . . the n -th order determinant $A = |a_{ij}|_n$ is the sum of all terms of the form $(-1)^x a_{nj_n} \dots$ " which (like some other equations and statements in the book) is a poetically distant view of the truth rather than a precise and detailed description. If the author could only rephrase such explanations in an unambiguous and accurate way, this would become a most useful book.

CEDRIC A. B. SMITH

Definitive Cell Biology

The Biology of the Cell Cycle. By J. M. Mitchison. Pp. v+313. (Cambridge University Press: London, December 1971.) £4.60 cloth; £1.60 paper.

PROFESSOR MITCHISON is one of the foremost analysts of the cell cycle and this book is the distillation of many years of thought and experiment in this field. It is a definitive work without any serious present competitor and unlikely to be surpassed in the foreseeable future. There are probably few scholars able to deal with so wide a range of material and fewer still able to deal with it critically.

The first two sections deal with methods for the study of the cell cycle: single cell techniques and synchronous cultures. The range and power of the various techniques are accurately discussed, and so are the uncertainties of interpretation introduced by the techniques themselves. It is difficult to think of any other source where the methodology for the study of the cell cycle is so succinctly and intelligently discussed. Two sections then follow dealing with DNA synthesis in a wide range of experimental material, from prokaryotes to higher animals. These sections may well be the most satisfactory ones, at least for those readers who are troubled by continuing uncertainty. For while we are still unsure of the enzymic basis of DNA replication and have no idea at all of the chemical nature of the signals that initiate and terminate it, we none the less do have a solid body of reproducible experiment on the geography and timing of DNA synthesis at various levels of cellular organization. This body of material Mitchison ably reviews. There is a single section on RNA synthesis, and, in some respects, this is the least satisfactory part of the book. For this the author can hardly be blamed; for while, over the years, experiments on DNA synthesis have become increasingly precise and thoughtful, experiments on RNA synthesis in whole cells are still very often difficult to interpret, and many experimenters in this field, despite prolonged instruction, remain incorrigibly silly. To the section on cell growth and protein synthesis and that on enzyme synthesis Mitchison brings an authority derived from his own experimental experience. He has perhaps done more than any other single worker to give precision to experiments on cell growth, and his recent work on periodic fluctuations in enzyme activity during the cell cycle has raised questions which may prove to be of fundamental importance to our understanding of how the synthesis of proteins really is regulated. In these two sections the personal commitment of the writer shows through, and it enlivens what might otherwise be very difficult subjects to dis-

cuss in an interesting way. The book ends with a collection of observations about cell organelles, respiration and pools and a thoughtful discussion on the control of cell division.

There is no doubt that this work will be of the greatest value to all those interested in cell biology; and it will be indispensable to those who have to teach the subject. The book is well produced, well indexed and contains, in addition to the text, about a thousand references carefully chosen and accurately documented.

HENRY HARRIS

Aardvark to Zebra

East African Mammals: An Atlas of Evolution in Africa. By Jonathan Kingdon. Pp. x+446. (Academic: New York and London, July 1971.) £12; \$35.

THE mammals of East Africa have for long been a focus of attention. The astonishing abundance and diversity of large mammals attracted first the hunter and more recently the ecologist and the tourist, while the attentions of naturalists and of veterinary and medical workers have resulted in the equally diverse smaller mammals being better known than those of most parts of the tropics. But among the long sequence of literature that has ensued there has never been any attempt at a comprehensive synthesis. For the resident biologist or the visiting naturalist alike there is virtually nothing available between the very superficial treatment of the field guide (and that only very recently) and the highly fragmentary confusion of papers on detailed taxonomic or ecological studies.

This volume is the first of three that are planned to cover all species of East African mammals. It deals with the primates and a number of small orders, namely the hyraxes, the pangolins, the aardvark and the dugong. The author makes it clear that the style and approach of each volume will vary—the second will contain the smaller mammals with emphasis on distribution and taxonomy, the third the larger species with special reference to their potential for exploitation. This first volume includes only thirty-two of the 360 or so species involved. This alone will dictate a very different approach to the remainder if the work is to be kept within bounds.

The introductory chapters include very useful and relevant accounts of the East African environment, with special emphasis on vegetation; and of the history and zoogeography of the East African mammal fauna. An appendix on the anatomy of mammals (dealing only with the skeletal and muscular systems) is less relevant but reflects the approach of the author throughout the following systematic section which is liberally illustrated with drawings of skeletons and of superficial dissections.

The latter in particular stem from the author's approach as an artist and are presumably intended to illustrate the evolutionary theme inherent in the title. But there is little correlation here between illustration and text which does not deal with the characters illustrated, while those aspects of structure that are more immediately relevant in terms of adaptation to the local environment, such as teeth and alimentary anatomy, are not illustrated.

In addition to these anatomical figures each species is illustrated by a beautifully executed black-and-white drawing supplemented in most cases by numerous "sketches from life". Four coloured plates depict very beautifully the intriguing array of pattern and colours in *Colobus* and *Cercopithecus*, although again one of these represents a diversion, dealing with a West African group. Distribution maps for each species are a very valuable feature and provide a sound and objective basis for zoogeographical analysis, an exercise that is too often based on those highly selected data that happen to fit the arguments.

The text covers many aspects of each species including distribution, variation, ecology and behaviour, but without any subdivision. In view of the overall evolutionary approach it seems a curious and irritating omission that no details are given of the variability in the transition zones where the two subspecies (or species?) of baboons and of vervet monkeys meet, both of them very intriguing situations for the student of speciation.

On the one hand, this volume must be welcomed as the only available synthesis of data on the mammals of East Africa, and a very valuable one at that, containing a wealth of personal experience of the subject. On the other hand, one must regret that the addition of a great deal of material of very marginal relevance to the main issue has enlarged the work to an extent and a price that will gravely limit its use, and that the data are not better organized for ease of retrieval and reference. In many respects it is a luxury but one that many will enjoy.

G. B. CORBET

Death of the Whale

The Blue Whale. By George L. Small. Pp. xiii+248. (19 plates.) (Columbia University: New York and London, June 1971.) \$9.95.

THE largest animal that has ever lived on this planet is virtually extinct. It is common knowledge that the blue whale has been hunted to this sorry state by that rapacious predator, man. This situation developed because the International Whaling Commission has no real power; and this impotence stems from the fact that the resources of the

high seas, including the whales, belong to nobody.

Conservationists, when they turn their attention from giant pandas and Javan rhinos to the plight of the whales—and this is usually once a year when the International Whaling Commission meets—confuse common property resources with those, like the whales, which are extra-territorial resources. This is the crux of the matter, for in this fact lies the failure of all the efforts both inside and outside the International Whaling Commission to save the whales, and until the publication of Professor Small's book, which in my opinion is the best book ever written on the blue whales and the whaling industry, this confusion has been perpetuated in most of the literature on the subject.

Professor Small's book is a case history which is more than informative, it is courageous. The author holds no punches: the failure of the International Whaling Commission to control the slaughter is explained and each country's part in the massacre is well documented, but he gives credit where it is due for efforts to conserve the whale stocks. This failure to manage the stocks of whales, he points out, is not just a matter of losing yet another species of animal. It is also the loss to a protein-hungry world of a valuable source of food. If the blue whale populations had been kept at an optimum level, around 60,000 individuals, they could have supplied mankind with a sustainable yield of some 6,000 blue whales a year, which would have provided over 105,000 tons of oil or enough to supply 2.5 ounces of margarine every day for a year to over 4 million human beings, or enough meat to supply a 6-ounce steak every day for a year to more than 3 million people.

The story of modern whaling is a tale, as this book forcibly illustrates, of miscalculation, short-sightedness and disregard for scientific evidence. It should have been comparatively easy to control the cropping of whales and to have implemented a plan that would have ensured the continuance of this important source of protein.

Conservation has come too late for the blue whale, but perhaps Professor Small's book, history as it is, may be taken as a warning and a guide for future action. To save the fin, sei and any other species that is exploitable, including that once prolific species—the sperm—some means of protecting them on the high seas has to be devised. Professor Small advocates, for example, control under one of the United Nations agencies. A good case for a new ocean regime under which all oceanic resources would be protected and exploited only under strict international control has been made. The

final point of the book, however, is made in the form of a question—in the author's words: "The tragedy of the blue whale is the reflexion of an even greater one, that of man himself. What is the nature of a species that knowingly and without good reason exterminates another?" ARTHUR BOURNE

Prebiology

Origins of Life. Edited by Lynn Margulis. (Proceedings of the first conference held at Princeton, New Jersey, May 21–24, 1967.) Pp. xiii+376. (Gordon and Breach: New York, London and Paris, July 1971.) (Published for the Interdisciplinary Communications Associates; Inc.) \$29.50; £12.30.

I CONFESS I opened this book with misgiving. For one thing, it is the eighth book on the same topic to be advertised as appearing in 1971; for another, in terms of pence per page, it is by far the most expensive. But a very cursory examination shows that at least it differs from the others in structure. It is an almost verbatim account of a two-day discussion held nearly five years ago in Princeton. There were twenty-five participants, and most of them are well-known specialists. Lynn Margulis has done a good job as editor. The book can be read at a sitting, and it reads more like a play than a text. The actors interrupt each other, crack jokes with each other, get a little annoyed now and then, but by and large give each other enough room to develop a host of conflicting theories. The book has been produced on behalf of the "Interdisciplinary Communications Program", and if it were not for the long delay in its publication it could be hailed as the most important book of the year on its topic. No field is moving faster.

But even some five years after the conference, this book takes a place that none other can fill. It is in the cut and thrust of argument that new ideas grow, and every reader can begin to sense the barriers that prevent one disputant from quite taking a point made by another.

The book is divided into two main sections, each dealing with the proceedings of a single day. The first section is dedicated to the fossil record, and after a brief argument about the age of the Earth, there follows an interesting debate about the primitive atmosphere. Stanley L. Miller leads off with a critical appraisal of what he calls the standard dogmas, and Raymond Siever refers to Sillén's thermodynamic arguments. It is good to hear Philip Abelson spell out his objections to assumptions made about the pH of the Precambrian ocean, and before long both Siever and Abelson have produced rival models for the origin of the Earth and its atmosphere. Siever postulates an early outgassing episode—and refers to it as the "Big

Burp hypothesis". Abelson prefers an Earth which accumulated from small pieces, with no atmosphere. Outgassing then produced water, nitrogen coming much later. Carbon dioxide, he claims, also lagged behind water, and hence the relative lack of carbonates in early Precambrian deposits. E. S. Barghoorn and J. W. Schopf continue with an invigorating discussion on the Gunflint, Fig Tree and Bitter Springs fossil localities, and Sidney W. Fox concludes this section by comparing some of these fossils with "proteinoid microspheres" that he has synthesized in the laboratory.

The second half of the book is devoted to a discussion of prebiological experimentation. Leslie E. Orgel and Miller discuss purines and pyrimidines, and Cyril Ponnamperna details his work on amino-acids. Polymerization mechanisms and the role of phosphates bring the arguments back to the environment, and more particularly to the conflicting claims advanced by Miller in favour of the oceans and Fox in favour of volcanic pools. This leads into a subsidiary argument: which were the first to polymerize, amino-acids or polynucleotides? Fox gives a very lucid account of his work on thermal "proteinoids", and Alexander Rich and Geoffrey Eglinton discuss how mixtures of purines and pyrimidines can carry genetic information even without polymerization. Carl Sagan and H. H. Pattee enter the argument and speculate on the origin of the genetic code. Soon most other members are drawn into the debate, which widens to include self-replication and the part played by the first membrane. Were there ever naked genes, or was there first a cell? Were Fox's microspheres involved, or did the genetic system come first and the membrane system later, as suggested by Sol Kramer and supported by most of the other biologists? Rich finally develops a theory for the evolution of the ribosomal system. Although the discussion is all the time getting more and more speculative, the speculation is far from idle. There is a constant reference to experiment, and an insistent inquisition.

In spite of its shortcomings, then, this is a healthy book to read, and a very useful companion to the hordes of other symposia that are now flooding the market. No student can hope to further the study of the origin of life if he follows the dogmas held by his peers. He must learn to query every experiment and every conclusion. This book will furnish him with an armoury of questions. We are told that it reports the first of five yearly conferences. We must surely encourage the publication of its successors, but next time let us have a cheap paperback that can be read by thousands rather than by hundreds—and let us have it soon!

P. C. SYLVESTER-BRADLEY

CORRESPONDENCE

Neptune Scandal

SIR,—Shame on you. One of the great stories in all of the history of science—that of the discovery of Neptune—has been mangled in your pages. In a report of the recent transfer of the 28-inch refracting telescope from Herstmonceux to Greenwich (*Nature*, **233**, 515; 1971) we read that initially the

“demand for the telescope arose through the so-called Neptune scandal: mathematicians in France and Cambridge independently predicted the existence of Neptune and the planet was observed by French astronomers, but there was no instrument in England capable of detecting it”.

The mathematicians were Urbain Jean Joseph LeVerrier (1811–1877), then Astronomer at the École Polytechnique in Paris, and John Couch Adams (1819–1892), then a fellow at St John's College. Accounts of their independent, brilliant calculations to locate an exterior planet to explain the observed behaviour of Uranus are found, for example, in Gould¹, Jones² and Grosser³.

Of course, the discovery was made by German astronomers (Johann Gottfried Galle and Heinrich Louis d'Arrest) after the planet was observed by James Challis at Cambridge. Jones² gives the following concise accounts of events in Berlin and in England.

“For on 23 September (1846) Galle, Astronomer at the Berlin Observatory, had received a letter from Le Verrier suggesting that he should search for the unknown planet, which would probably be easily distinguished by a disk. D'Arrest, a keen young volunteer at the Observatory, asked to share in the search, and suggested to Galle that it might be worth looking among the star charts of the Berlin Academy. . . . Galle took his place at the telescope, describing the configurations of the stars he saw, while d'Arrest followed them on the map, until Galle said: ‘And then there is a star of the 8th magnitude in such and such a position’, whereupon d'Arrest exclaimed: ‘That star is not on the map’. An observation the following night showed that the object had changed its position and proved that it was the planet.”

“Airy (Astronomer Royal) considered that the most suitable telescope with which to make the search for the new planet was the Northumberland telescope of the Cam-

bridge Observatory, which was larger than any telescope at Greenwich and more likely to detect a planet whose light might be feeble. . . .

“Challis decided to prosecute the search himself and began observing on 29 July, 1846, three weeks before opposition. The method adopted was to make three sweeps over the area to be searched, mapping the positions of all the stars observed, and completing each sweep before beginning the next. If the planet was observed it would be revealed, when the different sweeps were compared, by its motion relative to the stars.

“What followed was not very creditable to Challis. He started by observing in the region indicated by Adams: the first four nights on which observations were made were 29 July, 30 July, 4 August and 12 August. But no comparison was made, as the search proceeded, between the observations on different nights. He did indeed make a partial comparison between nights of 30 July and 12 August merely to assure himself that the method of observation was adequate. He stopped short at No. 39 of the stars observed on 12 August; as he found that all these had been observed on 30 July, he felt satisfied about the method of observation. If he had continued the comparison for another ten stars he would have found that a star of the 8th magnitude observed on 12 August was missing in the series of 30 July. This was the planet: it had wandered into the zone between the two dates.”

Challis had also observed the planet on August 4².

Yours faithfully,

BENEE F. SWINDEL

US Forest Service,
North Carolina State University,
Raleigh, North Carolina 27607

¹ Gould, Benjamin Apthorp, *Report to the Smithsonian Institution on the History of the Discovery of Neptune* (Smithsonian Institution, Washington, 1850).

² Jones, Harold Spencer, in *The World of Mathematics* (edit. by Newman, James R.), 822 (Simon and Schuster, New York, 1956).

³ Grosser, Morton, *The Discovery of Neptune* (Harvard University Press, Cambridge, Massachusetts, 1962).

“Blueprint for Survival”

SIR,—As professional biologists we wish to record our strong disapproval of your attitude to environmental issues as ex-

pressed in recent editorials, and in particular your derisory criticism of *The Ecologist's* “Blueprint for Survival” (*Nature*, **235**, 63; 1972). It is now widely acknowledged that the increasing material demands of a burgeoning world population are adversely affecting our environment and have grave implications for the future. It therefore seems laudable that a group of individuals should seek to identify the nature of the problem and to make recommendations for its solution. Such discussions are inevitably tentative but they surely claim to be more than a platform for wider debate.

We therefore support the request of Manning and Woodell (*Nature*, **235**, 179; 1972) calling for the evidence which leads you to dismiss the arguments in “Blueprint for Survival” and we would welcome a more constructive discussion of these problems in your columns.

Yours faithfully,

BOB ABEL	M. P. HARRIS
J. M. ANDERSON	A. K. KEPLER
H. R. ANDERSON	C. B. KEPLER
N. BAWDEN	CLARE LLOYD
GRAHAM BELL	DESMOND MORRIS
IAN CAMPBELL	C. M. PERRINS
C. J. CODY	R. P. PRYS-JONES
L. CORNWALLIS	B. A. C. SHAFFER
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RICHARD DAWKINS	J. N. M. SMITH
DAVID DAWSON	KATHY OATES
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HAMILTON	D. H. JANZEN
E. K. DUNN	GRAHAM HIRONS
P. GRANT	J. C. PERNETTA
T. R. HALLIDAY	T. G. O'DONNELL
PAUL HANDFORD	

University of Oxford

Coincident Congresses

SIR,—In 1972 the Eighth International Congress of Clinical Chemistry is being held in Copenhagen, June 19–23, and the Fourth International Congress of Endocrinology is being held in Washington, June 18–23.

Is it not one of the functions of ICSU and CIOMS to prevent such an unfortunate concurrence?

Yours faithfully,

D. N. BARON

Department of Chemical Pathology,
Royal Free Hospital
School of Medicine,
London

Publishing Ethics

SIR,—The suggestion that authors should regard it as unethical to negotiate with more than one book publisher (*Nature*, **234**, 367; 1971) is curious. Several unethical practices by major publishers overshadow the sort of calousness mentioned. Baumol and Heim (*Amer. Assoc. Univ. Prof. Bull.*, **53**, 30; 1967) give some examples as well as advice (see also *ibid.*, **53**, 275; and **55**, 121).

In addition to the usual risks an author runs of incompetent or biased referees (one recent paper of mine was rejected by a journal because two referees thought reduction required necessary as well as sufficient conditions), book publishers present other hazards, even legitimate ones. Contract requirements differ and cannot usually be known in advance. Acceptance or rejection of a manuscript may depend crucially on factors other than its quality. Delay in publication is a major variable. And so on.

An appropriate procedure is to send an abstract and chapter titles to several publishers to see if they are interested, assuming quality of the full manuscript to be adequate. Each publisher should know it is not the only one approached.

Much can be done at this stage with relatively little bother, and full negotiations can then be pursued with the most promising publisher.

Yours faithfully,

LEIGH VAN VALEN

*Committee on Evolutionary Biology,
University of Chicago,
5734 Ellis Avenue,
Chicago, Illinois 60637*

Case against Hysteria

SIR,—Whilst always treating the utterances of prophets of doom with due caution, I must nevertheless take issue with your editorial "The Case against Hysteria" (*Nature*, **235**, 63; 1972). The tone was complacent and patronizing (which helps nobody) and the content contains at least one point for rebuttal. For example, in the passage on power supply and oil resources, you state—and I agree—that nuclear power generation will become more competitive with (and, one assumes, will eventually replace) conventionally fuelled generation; but the point is that increasing supplies of power from whatever source will increase pollution, even if it is only

thermal pollution. Furthermore, the case of nuclear power was a particularly bad one for you to quote as the long-lived radioactive wastes represent a severe long term hazard. Fusion reactors which should produce no long-lived radioactive waste are much further off in time.

It can be argued that all this is perfectly obvious, that science/technology will produce solutions to environmental problems and that I am suffering from what Sir Peter Medawar called a "failure of nerve". However, experience shows that the operating time scales of both industry and government are far too long even when confronted with the obvious, so you must forgive me if I begin to feel distinctly uncomfortable.

The scramble for limited resources may have disastrous results (Korea and tungsten is surely a good enough warning) and it is right and proper to be concerned, even though this concern takes the form of "mere speculation" as you put it. Incidentally, since when has "speculation" been dismissed as "mere"?

Yours faithfully,

BRYAN LEVENE

*57A Burrard Road,
London NW6*

Announcements

University News

Dr S. J. Goldsack has been appointed to the new Control Data chair of computing science in the Department of Computing and Control, Imperial College, University of London.

Professor Kenneth Simkiss, Queen Mary College, London, has been appointed professor of zoology in the University of Reading.

Miscellaneous

Sir Hugh Linstead, chairman of Macarthy's Pharmaceuticals Limited, has been awarded the charter gold medal of the **Pharmaceutical Society of Great Britain**, for his outstanding services in promoting the interests of pharmacy. The Society's charter silver medal has been awarded to **Mr Cyril Turner**.

The platinum medal of the **Institute of Metals** has been awarded to the **Earl of Verulam**, Delta Metal Group, for his services to the non-ferrous metal industries. The Institute has awarded the Rosenhain medal to **Professor R. E. Smallman**, University of Birmingham, the W. H. A. Robertson medal and premium to **Mr D. Green**, UKAEA Reactor Fuel Element Laboratories, and the W. J.

Kroll medal and prize to **Dr O. Kubaschewski**, National Physical Laboratory. The British Committee of Award announces the following appointments to Harkness fellowships of the Commonwealth Fund: **A. H. Curran**, St John's College, Cambridge, chemistry; **I. S. Duff**, New College, Oxford, computer science; **P. Lennie**, King's College, Cambridge, neurophysiology; **I. R. McDougall**, Glasgow, nuclear medicine; **H. Pearson**, University College, London, chemistry; **R. B. Vinter**, St John's College, Cambridge, electrical engineering.

International Meetings

March 1, **Aspects of Quality Control during Manufacturing**, Hoddesdon (Society for Analytical Chemistry, 9-10 Savile Row, London W1X 1AF).

March 7, **The Role of Metals in Biological Systems, excluding Enzymes**, London (P. Woodward, School of Chemistry, The University, Bristol BS8 1TS).

March 8, **Adhesives**, Birmingham (The Secretary, Institution of the Rubber Industry, 4 Kensington Palace Gardens, London W8 4QR).

March 8, **Is There a Need for Impurity Standards in Organic Analysis?**, London (Society for Analytical Chemistry, 9-10 Savile Row, London W1X 1AF).

March 8, **Present Trends in Analytical**

Research, York (Society for Analytical Chemistry, 9-10 Savile Row, London W1X 1AF).

March 8-9, **Climatic Resources and Economic Activity**, Aberystwyth (J. A. Taylor, Department of Geography, University College of Wales, Llandinam Building, Penglais, Aberystwyth, Cardiganshire).

March 8-10, **Molecular Studies in Viral Neoplasia**, Houston (Symposium Registration, University of Texas at Houston, M. D. Anderson Hospital and Tumor Institute, Houston, Texas 77025, USA).

March 9, **Chemical Engineering Developments in Mineral Processing**, London (J. R. Theobald, Atomic Weapons Research Establishment, Building A.75, Aldermaston, Reading RG7 4PR).

March 13-16, **Physics Exhibition**, London (Institute of Physics, 47 Belgrave Square, London SW1X 8QX).

March 14, **Survival and the Industrial Librarian**, London (Miss P. Barrie, IBM (UK Laboratories) Ltd, Hursley Park, Winchester, Hampshire).

March 16, **Developing and Marketing a New Food Product**, London (Institute of Food Technologists, c/o Cotswold House, Westhorpe, Sibbertoft, Leicester LE16 9UL).

March 16, **Carbon in Effluents**, Nottingham (Society for Analytical Chemistry, 9-10 Savile Row, London W1X 1AF).

March 16–17, **The Prevention of Abuse of the Oceans**, London (Council of Engineering Institutions, 2 Little Smith Street, Westminster, London SW1).

March 20, **Link Scheme for the Electronics Industry**, London (Secretary, IEE, Savoy Place, London WC2).

March 20–21, **Semiconductor Injection Lasers and their Applications**, Cardiff (Mr D. R. Hub, Short Courses Section, UWIST, King Edward VII Avenue, Cardiff CF1 3NU).

March 22–23, **Clean Air**, Manchester (National Society for Clean Air, 134–137 North Street, Brighton BN1 1RG).

March 23, **Food Emulsions**, London (Food Group, Society of Chemical Industry, 14 Belgrave Square, London SW1).

March 23–24, **Recent Advances in General Analytical Chemistry**, Glasgow (Society for Analytical Chemistry, 9–10 Savile Row, London W1X 1AF).

March 23–26, **Studies in Building Conservation**, London and Venice (Centre for Advanced Studies in Environment, The Architectural Association, 36 Bedford Square, London WC1B 3ES).

March 24, **Safety in the Chemical Laboratory**, Chepstow (Society for Analytical Chemistry, 9–10 Savile Row, London W1X 1AF).

March 27–28, **British National Committee for High Speed Photography Conference**, Egham, Surrey (C. W. Husbands, Central Unit for Scientific Photography, Royal Aircraft Establishment, Farnborough, Hampshire).

March 27–28, **Combustion-generated Pollution and its Control**, Birmingham (Professor C. F. Cullis, Department of Chemistry, The City University, St John Street, London EC1).

March 27–29, **Biosynthesis and its Control in Plants**, Canterbury (Dr B. V. Milborrow, Milstead Laboratory, Broad Oak Road, Sittingbourne, Kent).

March 28, **Fibre Optical Communications**, London (Institute of Physics, 47 Belgrave Square, London SW1X 8QX).

March 28–29, **Potable Water from Polluted Sources**, London (Society for Water Treatment and Examination, 69 Disraeli Crescent, High Wycombe, Buckinghamshire).

April 4–8, **British Mathematical Colloquium**, Glasgow (Dr I. Anderson, Department of Mathematics, University Gardens, Glasgow G12 8QL).

April 5–7, **Jet Cutting Technology**, Warwick (The Organizing Secretary, First ISJCT, BHRA Fluid Engineering, Cranfield, Bedford).

April 5–7, **Phase Analysis: Identification and Quantitative Determination**, Hull (Dr W. J. Duffin, Department of Physics, University of Hull, Hull HU6 7RX).

April 6, **Bioavailability of Drugs from Medicines**, London (Society for Drug Research, c/o Mrs C. Bates, Chelsea College, Manresa Road, London SW3).

April 6, **Statistical Methods for Chemists**, London (Assistant Secretary, Society of Chemical Industry, 14 Belgrave Square, London SW1).

April 6–7, **Diffusion in Polymeric Systems**, Aberdeen (Dr J. B. Craig, Department of Chemistry, University of Aberdeen, Meston Walk, Old Aberdeen AB9 2UE).

April 9–12, **Noise in an Industrial Environment**, Guildford (Meetings Officer, Institute of Physics, 47 Belgrave Square, London SW1X 8QX).

April 10–12, **Noise and Vibration Control for Industrialists**, Bath (Society of Environmental Engineers, 68a Wigmore Street, London W1H 9DL).

April 10–12, **Skin-Environmental Responses and Protection**, Oxford (The General Secretary, Society of Cosmetic Chemists of Great Britain, 56 Kingsway, London WC2).

April 10–13, **Interfacial and Surface Phenomena**, York (Meetings Officer, Institute of Physics, 47 Belgrave Square, London SW1).

April 10–14, **Spices**, London (Tropical Products Institute, 56–62 Gray's Inn Road, London WC1X 8LU).

April 10–14, **Chemical Society Inaugural Meeting**, Manchester (The Chemical Society, Burlington House, London W1V 0BN).

April 11–12, **The Commercial Utilization of Seaweeds**, Southsea (R. E. Marshall, The Pharmaceutical Society of Great Britain, 17 Bloomsbury Square, London WC1A 2NN).

April 11–13, **Elastohydrodynamic Lubrication**, Leeds (Conference Publicity Section, Institution of Mechanical Engineers, 1 Birdcage Walk, London SW1).

April 11–13, **Industrial Measurement and Control by Radiation Techniques**, Guildford (Manager, Conference Department, IEE, Savoy Place, London WC2R 0BL).

April 11–13, **Interdisciplinary Investigation of the Brain**, Oxford (Dr J. P. Nicholson, Physics Department, Westminster Hospital, Page Street Wing, London SW1).

April 11–13, **Management and Information**, Manchester (D. F. Styles, 55 Penrhyn Avenue, Middleton, Manchester M24 1FP).

April 11–13, **Technologists for Metallurgical Industries**, Scarborough (The Meetings Secretary, The Institution of Metallurgists, Northway House, High Road, Whetstone, London N20 9LW).

April 12, **Frictional Materials**, Uxbridge (I. B. Smith, Department of Polymer Science and Technology, Brunel University, Kingston Lane, Uxbridge, Middlesex).

April 12–14, **Machine Perception of Patterns and Pictures**, Teddington (Meetings Assistant, Institute of Physics, 47 Belgrave Square, London SW1).

April 12–14, **Parameters of the Vascular System relating to Radiation Response**, London (Professor P. J. Lindop, Department of Radiobiology, Medical College of St Bartholomew's Hospital, Charterhouse Square, London EC1M 6BQ).

April 13–14, **Industrial Crystallization**, London (Dr D. Elwell, Physics Department, Portsmouth Polytechnic, Park Road, Portsmouth).

April 14, **The Patronage of Science in the Nineteenth Century**, London (Assistant Secretary, British Society for the History of Science, 47 Belgrave Square, London SW1).

April 17–18, **Calorimetry in Biological Sciences**, Birmingham (Dr J. M. Creeth, Lister Institute of Preventive Medicine, Chelsea Bridge Road, London SW1).

April 17–18, **Drug Receptors**, London (Miss G. Blunt, c/o Department of Pharmacology, University College, Gower Street, London WC1E 6BT).

April 17–18, **The Scientific and Technological Gap in Latin America**, Lincoln, Nebraska (Dr R. Esquenazi-Mayo, University Extension Division, University of Nebraska-Lincoln, Lincoln, Nebraska 68508, USA).

April 17–19, **The Electrical Double Layer**, Bristol (Dr R. Parsons, Department of Physical Chemistry, The University, Bristol BS8 1TS).

April 17–19, **Ultra High Vacuum**, Swansea (Meetings Officer, Institute of Physics, 47 Belgrave Square, London SW1).

April 17–20, **Atomic and Molecular Physics**, Brighton (Meetings Officer, Institute of Physics, 47 Belgrave Square, London SW1).

April 17–20, **Geochemical Exploration**, London (The Secretary, Institution of Mining and Metallurgy, 44 Portland Place, London W1N 4BR).

April 17–21, **Service to Industry—Today and Tomorrow**, London (Paint Research Association, Waldegrave Road, Teddington, Middlesex).

April 18–19, **Electrostatics; Fundamentals, Applications and Hazards**, Southampton (Meetings Officer, Institute of Physics, 47 Belgrave Square, London SW1X 8QX).

April 18–19, **Mathematical and Hydraulic Modelling of Estuarine Pollution**, Stevenage (The Director, Water Pollution Research Laboratory, Elder Way, Stevenage, Hertfordshire).

April 18–20, **Nuclear Physics; Nuclear Structure and Nuclear Reactions**, Birmingham (Meetings Officer, Institute of Physics, 47 Belgrave Square, London SW1X 8QX).

Reports and Publications

not included in the Monthly Books Supplement

Great Britain and Ireland

Microscopes to the End of the Nineteenth Century. By F. W. Palmer and A. B. Sahar. (A Science Museum Illustrated Booklet.) (London: HMSO, 1971.) 35p net. [2911]

The British Council. Annual Report 1970/1971. Pp. 102. (London: The British Council, 1971.) [2911]

Science Research Council. The Royal Observatory, Edinburgh. Publications, Vol. 7, No. 6: Spectrophotometry of Cool Stars in the Near Infrared. II Results for a Region in the Direction of the Galactic Anticentre. By K. Nandy and F. Smiraglio. Pp. 73-83. (Edinburgh: The Royal Observatory, 1971.) 50p net. [2911]

Fluids in Motion. By Professor E. Markland. (An Inaugural Lecture delivered before The Queen's University of Belfast on 14 February 1968.) Pp. 14. (Belfast: The Queen's University, 1968.) 40p. [112]

The Royal Society. Report of Council for the year ended 31 August 1971. Pp. 99. (London: The Royal Society, 1971.) [112]

University of Nottingham. Report of the School of Agriculture, 1970-1971. Pp. 120. (Sutton Bonington, Loughborough: University of Nottingham School of Agriculture, 1971.) 60p. [112]

Images from Life. (Publication No. 689.) Pp. 110. (London: British Museum (Natural History), 1971.) [212]

Conversions of Buildings for Science and Technology. (Paper No. 5.) Pp. 36. (London: Laboratories Investigation Unit, Department of Education and Science, 1971.) gratis. [312]

The Royal Society. Scientific Research in Schools Committee—Report to Council 1971. Pp. 21. (London: The Royal Society, 1971.) [612]

Technological Innovation—a Methodology. By Dr. L. Bruce Archer. (SPF Special Publications Series.) Pp. 64. (London: Inforlink Ltd., 1971.) £4.50. [612]

Young Fabian Pamphlet No. 27: Our Children's Teachers Into the Seventies. By Isla Calder. Pp. 28. 30p. Fabian Research Series No. 298: Collective Bargaining and Inequality. By Jim Skinner. Pp. 20. (London: Fabian Society, 1971.) [612]

Chromatography in Clinical Biochemistry Using Flexible Thin Layers. By R. S. Ersser. Pp. 24. (Colnbrook, Bucks: Koch-Light Laboratories, Ltd., 1971.) [612]

Forestry Commission. Forest Record No. 78: Loading and Unloading Timber Lorries. By Arthur Sutton and T. R. Sawyer. Pp. 53. 30p net. Forest Record No. 79: Nothofagus Plantations in Great Britain. By M. Nimmo. Pp. 12. 17p net. Research and Development Paper No. 82: Fume Damage to Forests—Summaries of Papers presented to the International Symposium of Forest Fume Damage Experts, Essen, West Germany, September 1970. Pp. 50. Report on Forest Research 1971. Pp. vii + 171 + 12 plates. (London: HMSO, 1971.) [612]

Metropolitan Water Board. Forty-fourth Report on the Results of the Bacteriological, Chemical and Biological Examination of the London Waters for the years 1969-1970. By E. Windle Taylor. Pp. 163. (London: Metropolitan Water Board, 1971.) £2.50. [712]

Philosophical Transactions of the Royal Society of London. A: Mathematical and Physical Sciences. Vol. 270, No. 1207: Thermal Theory of Spontaneous Ignition—Criticality in Bodies of Arbitrary Shape. By T. Boddington, P. Gray and D. I. Harvey. Pp. 467-506. £1.05; \$2.75. Vol. 270, No. 1208: Measurements of the Viscometric Functions for a Fluid in Steady Shear Flows. By W. G. Pritchard. Pp. 507-556. £1.30; \$3.40. (London: The Royal Society, 1971.) [712]

ITV 2: a Submission to the Minister of Posts and Telecommunications by the Independent Television Authority, December 1971. Pp. 24. (London: ITA, 1971.) [812]

Bulletin of the British Museum (Natural History). Entomology. Vol. 26, No. 7: A Preliminary Revision of the Genus *Oxya* Audinet-Serville (Orthoptera: Acridoidea). By D. Hollis. Pp. 267-343. £2.90. Zoology. Vol. 21, No. 6: Lizards and Snakes from Transjordan, Recently Acquired by the British Museum (Natural History). By Y. L. Werner. Pp. 213-256 + 6 plates. £2. Vol. 21, No. 7: *Conchoecia* from the North Atlantic, the 'Proceras' Group. By Martin Vivian Angel. Pp. 257-283. £1.05. (London: British Museum (Natural History), 1971.) [812]

Underwater Handbook: Western Approaches to the British Isles—Oceanographic and Meteorological Data. (N.P. 625.) Pp. ix + 123 + 15 plates. (London: Hydrographic Department, Ministry of Defence, 1971.) £3.50 net. [812]

Symposium on Antarctic Ice and Water Masses, Tokyo, Japan, 19 September 1970. Edited by Sir George Deacon. Pp. x + 113. (Cambridge: Scientific Committee on Antarctic Research, 1971.) [812]

Genesis Four (On the Origin of Comets). By Douglas Rookes. Pp. 38. (London: Aridilla Books, 1971.) 50p. [912]

British Standards Institution. International Organization of Legal Metrology. Vocabulary of Legal Metrology—Fundamental Terms. Edition March 1969. Official OIML version in French—unofficial BSI translation in English. (PD 6461.) Pp. 180. (London: British Standards Institution, 1971.) £5. [912]

Other Countries

Université du Droit et de la Santé de Lille. Facultés et Unités d'Enseignement et de Recherche des Sciences Médicales. Appareil Respiratoire et Nuisances: Recherches Thématiques Coordonnées au Cours de la Dernière Décennie. Pp. 61. (Lille: Université du Droit et de la Santé de Lille, 1971.) [2211]

Organization for Economic Co-operation and Development. OECD Informatics Studies, No. 2: Digital Information and the Privacy Problem. By G. B. F. Niblett. Pp. 58. (Paris: OECD; London: HMSO, 1971.) 9 francs; 70p; \$2; 8 Sw. francs; 6.30 DM. [2311]

The Institute of Science, Bombay. Golden Jubilee Commemoration Volume. Pp. 232. (Bombay: The Institute of Science, 1971.) [2411]

United Nations: Department of Economic and Social Affairs. Strategy Statement on Action to Avert the Protein Crisis in the Developing Countries. (Report of the Panel of Experts on the Protein Problem Confronting Developing Countries, UN Headquarters, 3-7 May 1971.) Pp. viii + 27. (New York: United Nations, 1971.) \$1. [2411]

Australia: Commonwealth Scientific and Industrial Research Organization, Australia. CSIRO Twenty-third Annual Report, 1970/71. Pp. 98. (East Melbourne, Victoria: CSIRO, 1971.) [2411]

Smithsonian Contributions to Paleobiology, No. 6: Functional Morphology and Biofacies Distribution of Cheilostome Bryozoa in the Danian Stage (Paleocene) of Southern Scandinavia. By Alan H. Cheetham. Pp. iv + 87 (17 plates). (Washington, DC: Smithsonian Institution Press, 1971. For sale by US Government Printing Office.) \$1.50. [2611]

Association for the Aid of Crippled Children. Annual Report 1970/1971. Pp. 63. (New York: Association for the Aid of Crippled Children, 1971.) [2611]

Coronary Heart Disease—Report of a Committee of the Royal Society of New Zealand. Pp. 98. (Wellington: The Royal Society of New Zealand, 1971.) [2911]

Bulletin of the American Museum of Natural History, Vol. 145, Article 1: Systematics and

Behavior of Some North American Woodpeckers. Genus *Picoides* (Aves). By Lester L. Short. Pp. 1-118. (New York: American Museum of Natural History, 1971.) \$3.60. [2911]

Smithsonian Contributions to Zoology. No. 85: Systematics, Distribution and Evolution of the Chub Genus *Nocomis* Girard (Pisces, Cyprinidae) of Eastern United States, with descriptions of New Species. By Ernest A. Lachner and Robert E. Jenkins. Pp. iii + 97. \$1.25. No. 95: Neotropical Microlepidoptera XIX—Notes on and New Species of Oecophoridae (Lepidoptera). By J. F. Gates Clarke. Pp. 39. \$0.50. (Washington, DC: Smithsonian Institution Press, 1971. For sale by US Government Printing Office.) [2911]

US Department of the Interior: Geological Survey. Professional Paper 486-E: Salinity of Surface Water in the Lower Colorado River—Salton Sea Area. By B. Ireland. Pp. iv + 40. \$0.50. Professional Paper 657-A: Geology of the Van Buren and Lavaca Quadrangles, Arkansas and Oklahoma. By Boyd R. Haley and Thomas A. Hendricks. Pp. iv + 41 + 7 plates. (Washington, DC: Government Printing Office, 1971.) [2911]

Publications of the United States National Museum (1947-1970). (Bulletin 298.) Pp. 77. (Washington, DC: Smithsonian Institution Press, 1971. For sale by US Government Printing Office.) \$0.60. [2911]

The Fine Creek and Mill River Watersheds, Fairfield, Connecticut: an Ecological Guide to Open Space Land Use. By Whitney Beals and Peter Westover. Pp. iv + 55. (New Haven, Conn.: School of Forestry, Yale University, 1971.) [2911]

Canada: Department of Energy, Mines and Resources. Geological Survey of Canada. Bulletin 175: Some Petrological Aspects of the Harbour Coal Seam, Sydney Coalfield, Nova Scotia. By A. R. Cameron. Pp. 74 (4 plates). \$2. Bulletin 189: Precambrian Fossils, Pseudofossils, and Problematica in Canada. By H. J. Hofmann. Pp. 146. \$3. Bulletin 210: Ordovician Trilobites from the Central Volcanic Mobile Belt at New World Island, North-eastern Newfoundland. By W. T. Dean. Pp. 37 + 7 plates. \$2. (Ottawa: Information Canada, 1971.) [2911]

Rubber Research Institute of Malaya. Annual Report 1970. Pp. 133. (Kuala Lumpur: Rubber Research Institute of Malaya, 1971.) \$3. [3011]

World Health Organization. International Standards for Drinking-Water. Third edition. Pp. 70. (Geneva: WHO; London: HMSO, 1971.) 9 Sw. francs; 90p; \$3. [3011]

Research Policy, Vol. 1, No. 1, November 1971. (A journal devoted to research policy, research management and planning.) Edited by C. Freeman, T. C. Sinclair, H. Krauch and R. Coenen. Pp. 1-102. Published quarterly. Subscription price per volume of about 400 pages is Dfls. 91.00. (Amsterdam: North-Holland Publishing Company, 1971.) [312]

Australia: Commonwealth Scientific and Industrial Research Organization. Annual Report of the Division of Irrigation Research 1970-71. Pp. iv + 78. (Griffith, NSW: CSIRO, 1971.) [312]

Unesco Field Science Office for Africa, Nairobi. Survey on the Scientific and Technical Potential of the Countries of Africa/Enquête sur le Potentiel Scientifique et Technique des Pays D'Afrique. Pp. 209. (Paris: Unesco, 1970.) 24 francs; £1.80; \$6. [312]

Smithsonian Contributions to Zoology. No. 84: Butterflies of the Genus *Vanessa* and of the Resurrected Genera *Bassaris* and *Cynthia* (Lepidoptera: Nymphalidae). By William D. Field. Pp. 105. \$1.50. No. 91: Systematics, Distribution, and Evolution of the *Nocomis biguttatus* Species Group (Family Cyprinidae: Pisces) with a Description of a New Species from the Ozark Upland. By Ernest A. Lachner and Robert E. Jenkins. Pp. 28. \$0.40. (Washington, DC: Smithsonian Institution Press, 1971. For sale by US Government Printing Office.) [312]

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